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2nd Edition

M.C.G. Davies Morel



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Equine Reproductive Physiology, Breeding and Stud Management

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Dedication

This book is dedicated to my husband Roger,
as thanks for all his love and support.

Equine Reproductive Physiology, Breeding and Stud Management

2nd Edition

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1

The Reproductive Anatomy of the Mare

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1.1. Introduction

This chapter will deal with the reproductive anatomy of the mare, detailing the individual structures along with their function. Further accounts may be found in other texts, such as Ashdown and Done (1987), Frandson and Spurgen (1992), Ginther (1992), Kainer (1993), Dyce *et al.* (1996), Bone (1998), Senger (1999), Bergfelt (2000) and Hafez and Hafez (2000). The reproductive tract of the mare may be considered as a Y-shaped tubular organ with a series of constrictions along its length. The perineum, vulva, vagina and cervix can be considered as the outer protective structures, providing protection for the inner, more delicate structures, the uterus, Fallopian tubes and

ovaries, responsible for fertilization and embryo development. Figure 1.1, taken after slaughter, shows the reproductive structures of the mare, and Figs 1.2 and 1.3 illustrate these diagrammatically. Each of these structures will be dealt with in turn in the following account.

1.2. The Vulva

The vulva (Fig. 1.4) is the external area of the mare's reproductive system, protecting the entrance to the vagina. The outer area is pigmented skin with the normal sebaceous and sweat glands, along with the nerve and blood supply normally associated with the skin of the mare. The inner area is lined by

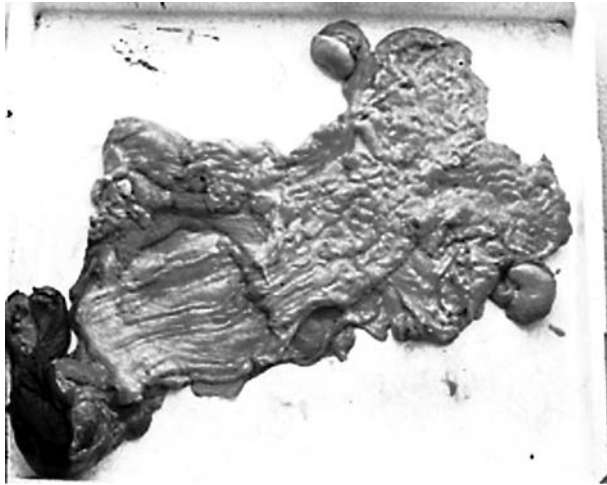


Fig. 1.1. The mare's reproductive tract after slaughter and dissection (see also Fig. 1.3).

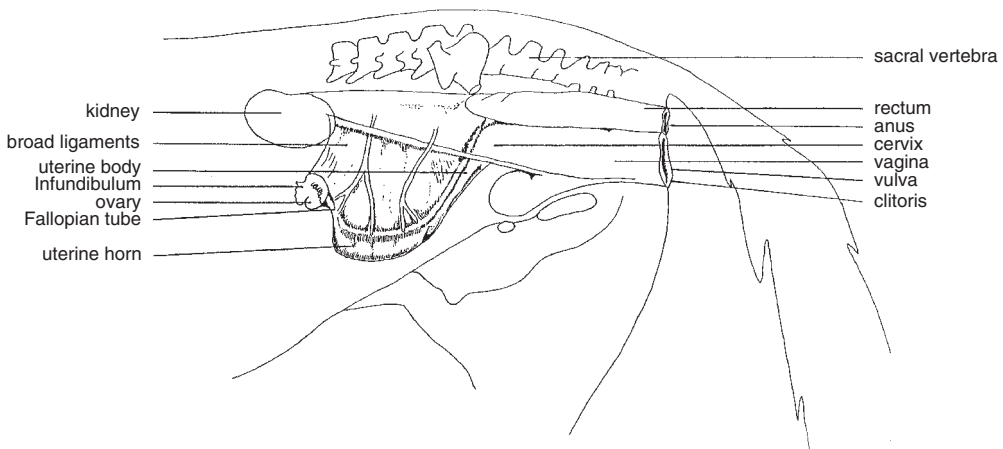


Fig. 1.2. A lateral view (from the side) of the mare's reproductive tract.

mucous membrane and is continuous with the vagina. The upper limit (the dorsal commissure) is situated approximately 7 cm below the anus. Below the entrance to the vagina, in the lower part of the vulva (the ventral commissure), lie the clitoris and the three sinuses (ventral, medial and lateral) associated with it. These sinuses are of importance in the mare as they provide an ideal environment for the harbouring of many venereal disease (VD) bacteria, such as *Taylorella equigenitalis* (causal agent for contagious equine metritis (CEM)), *Klebsiella pneu-*

moniae and *Pseudomonas aeruginosa*. Hence, this area is regularly swabbed in mares prior to covering and, indeed, in the Thoroughbred industry, such swabbing is compulsory (McAllister and Sack, 1990; Ginther, 1992; Horse Race Betting Levy Board, 2001). Within the walls of the vulva lies the vulva constrictor muscle, running either side of the length of the vulval lips. This muscle acts to maintain the vulval seal and to invert and expose the clitoral area during oestrus, known as winking (Ashdown and Done, 1987; Kainer, 1993).

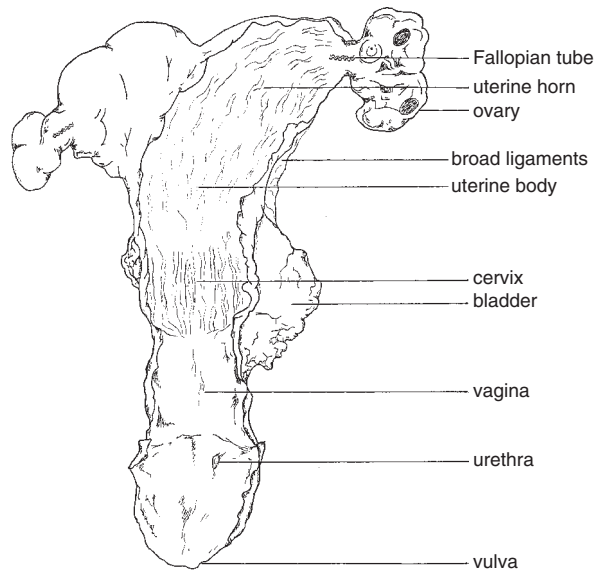


Fig. 1.3. The mare's reproductive tract: a diagrammatic representation of Fig. 1.1.



Fig. 1.4. The vulval area of the mare: in this instance the conformation of the perineal area is poor with the anus sunken cranially, opening the vulva up to faecal contamination.

1.3. The Perineum

The perineum is a rather loosely defined area in the mare, but includes the outer vulva and adjacent area, along with the anus and the surrounding area. In the mare the conformation of this area is of clinical importance, due to its role in the protection of the genital tract from the entrance of air. Malconformation in this area predisposes the mare to a condition termed pneumovagina, or vaginal wind-sucking, in which air is sucked in and out of the vagina through the open vulva. Along with this passage of air also go bacteria, which bombard the cervix, exposing it to unacceptably high levels of contamination, which it is often unable to cope with, especially during oestrus when it is less competent. Passage of bacteria into the higher, more susceptible parts of the mare's tract may result in bacterial infections, such as CEM, and other venereal diseases, leading to endometritis. Chapter 19 gives further details on the causes of VD infections in the mare, all of which adversely affect fertilization rates (Ginther, 1992; Easley, 1993; Kainer, 1993).

1.3.1. Protection of the genital tract

As mentioned previously, adequate protection of the genital tract is essential to prevent the adverse affects of pneumovagina. There are three seals within the tract: the vulval seal, the vestibular or vaginal seal and the cervix. These are illustrated in Fig. 1.5.

The perineal area plus the vulva constrictor muscle in the walls of the vulva form the vulval seal. The vestibular seal is formed by the natural apposition of the walls of the posterior vagina, where it sits above the floor of the pelvic girdle and the hymen, if still present. The tight muscle ring within the cervix forms the cervical seal. This series of seals is affected by the conformation of an individual and also by the stage of the oestrous cycle (Figs 1.6–1.8).

The ideal conformation is achieved if 80% of the vulva lies below the pelvic floor. A simple test can be used to assess this. If a sterile plastic tube is inserted through the vulva into the vagina and allowed to rest on the vagina floor, the amount of vulva lying below this tube should be approximately 80% in well-conformed mares. This technique is illustrated diagrammatically in Fig. 11.3.

If the ischium of the pelvis is too low, then the vulva tends to fall towards the horizontal plane, as seen in Fig. 1.6. This opens up the vulva to contamination by faeces, increasing the risk of uterine infection. Pascoe (1979a) suggested that mares should be allocated a Caslick index derived by multiplying the angle of inclination of the vulva with the distance from the ischium to the dorsal commissure. This index can then be used to classify mares into three types and so predict the likely occurrence of endometritis (Fig. 1.7).

The effect of poor conformation of the perineum area may be alleviated by a Caslick's vulvoplasty operation, developed by Dr Caslick in 1937 (Caslick, 1937). The lips on either side of the upper vulva are cut, and the two sides are then sutured together. The two raw edges heal together, as in the healing of an open wound, and hence seal the upper part of the vulva. The hole left at the ventral commissure is adequate for urination but prevents the passage of faeces into the vagina (Fig. 1.9).

The chance that a mare requiring a Caslick's operation will pass on the trait to her offspring is reasonably high. This, coupled with the fact that the operation site has to be cut to allow mating and foaling, casts doubt on whether such mares should be

Fig. 1.5. The seals of the mare's reproductive tract during dioestrus.

(a)

(b)

(c)

Fig. 1.6. The effect of conformation on the competence of the vulval, vestibular and cervical seals in the mare. (a) A low ischium (pelvic floor) results in an incompetent vestibular seal. In this case the vulval seal is still competent; therefore, infection risk is limited. (b) A low ischium results in an incompetent vestibular seal. In this case, the vulval seal is also incompetent; therefore, infection risk is increased. (c) An incompetent vestibular and vulval seal plus a sloping perineal area results in a significant infection danger, especially from faecal contamination.

bred. Mares that have been repeatedly cut and resutured become increasingly hard to perform a Caslick's operation on, as the lips of the vulva become progressively more fibrous and therefore difficult to suture. In such cases, a procedure termed a Pouret may be carried out (Pouret, 1982). This is a more major operation and involves the realignment of the anus as well as the vulva.

This perineal malformation is particularly prevalent in Thoroughbred mares. It is causal to both pneumovagina (collection of air within the vagina) and urinovagina (collection of urine within the vagina), both conditions being precursors for endometritis and hence infertility. The condition tends to be exacerbated in mares that lack condition and also in multiparous, aged mares. Its continued existence is largely due to the selection of horses for athletic performance rather

than reproductive competence (Caslick, 1937; Pascoe, 1979a; Pouret, 1982; Le Blanc, 1991; Easley, 1993).

The oestrous cycle also has an effect on the competence of the three seals. Further details of the effect of the oestrous cycle on the reproductive tract are detailed in Chapter 3. However, in summary, oestrus results in the slackening of all three seals, due to a relaxation of the muscles associated with the reproductive tract, especially the cervix (Fig. 1.8). This allows intromission at covering but also decreases the competence of the reproductive-tract seals and so increases the chance of bacterial invasion. In part this is compensated for by elevated oestradiol levels characteristic of oestrus, which enhance the mare's immunological response, thus reducing the chance of uterine infection, despite the increased chance of bacterial invasion.

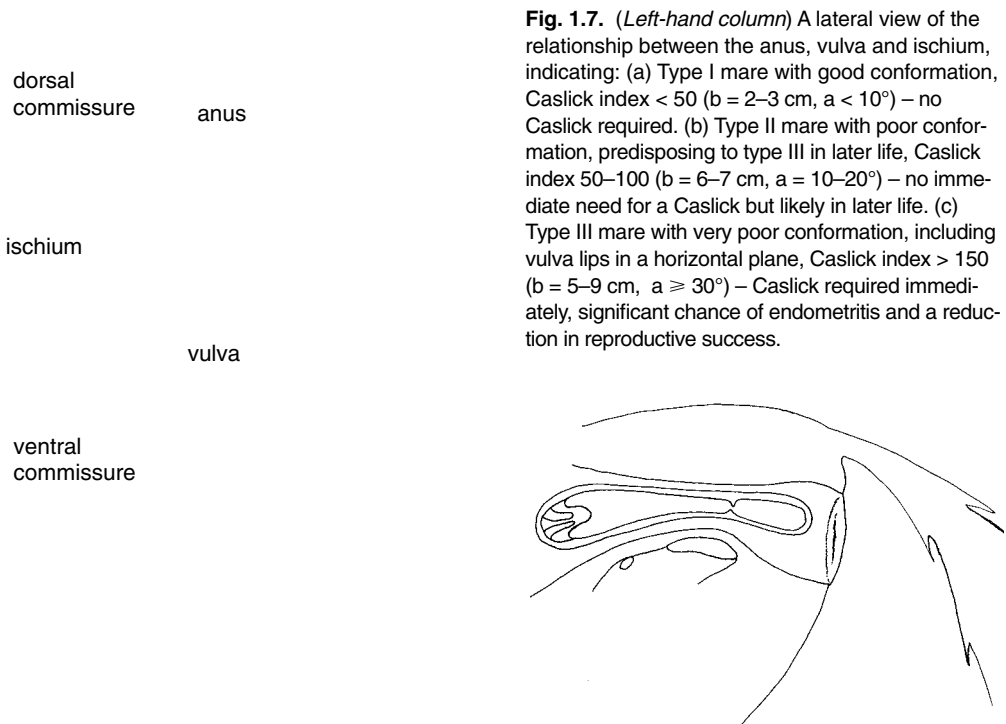


Fig. 1.8. The effect of oestrus on the competence of the vulval, vestibular and cervical seals in the mare: oestrus causes a relaxation of the seals and therefore an increase in infection danger.

1.4. The Vagina

The vagina of the mare is on average 18–23 cm long and 10–15 cm in diameter. In the well-conformed mare, the floor of the vagina should rest upon the ischium of the pelvis and the walls are normally collapsed and apposed, forming the vestibular seal. The hymen, if present, is also associated with this seal and divides the vagina into anterior (cranial) and posterior (caudal) sections. In some texts the posterior vagina is referred to as the vestibule. The urethra, from the bladder, opens just caudal to the hymen. Within the body cavity, the vagina is mainly covered by the peritoneum and is surrounded by loose connective tissue, fat and blood vessels. The walls of the vagina are muscular with a mucous lining; the elasticity conferred by the muscle layer allows the major stretching required at parturition (Fig. 1.10).



(a)



(b)



(c)

Fig. 1.9. A Caslick operation in the mare showing (a) the cutting of the vulval lips, (b) suturing and (c) the finished job.

The vagina acts as the first protector and cleaner of the system, containing acidic to neutral secretions, originating from the cervix, which are bactericidal. The vagina itself is aglandular, except just cranial to the vulval

lips, where secretory glands are located. These secretions, however, have the disadvantage of attacking the epithelial cell lining of the vagina, necessitating the protective mucous layer, and of being spermicidal. Thus, at ejacu-

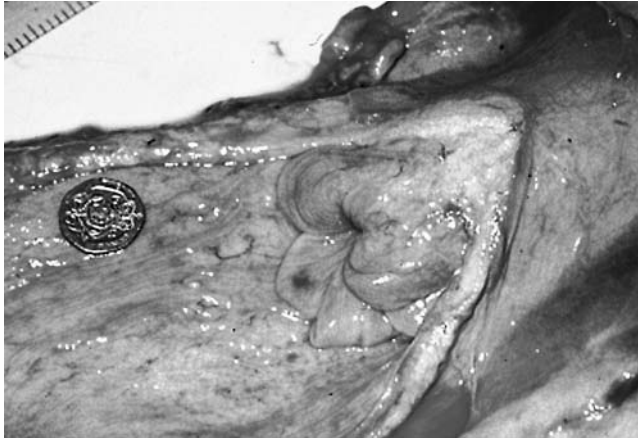


Fig. 1.10. The internal surface of the mare's vagina (the coin measures 21 mm in diameter).

lation, sperm are deposited into the top of the cervix and/or bottom of the uterus, to avoid the detrimental effect of the relatively high pH within the vagina. The exact composition of vaginal secretion is controlled by the cyclical hormonal changes of the mare's reproductive cycle (Ginther, 1992; Kainer, 1993).

1.5. The Cervix

The cervix lies at the entrance to the uterus. It is a tight, thick-walled, sphincter muscle, acting as the final protector of the system.

In the sexually inactive, dioestrous state, it is tightly contracted, white in colour and measures on average 6–8 cm long by 4–5 cm in diameter; cervical secretion is minimal and thick in consistency. The muscle tone and therefore cervix size, along with its secretion, are again governed by cyclic hormonal changes. Muscle tone relaxes during oestrus and there is an increase in secretion, easing the passage of the penis into the entrance of the cervix. The oestrous cervix appears pink in colour and may be seen protruding or 'flowering' into the vagina (Fig. 1.11; Lieux, 1970).



Fig. 1.11. The oestrus cervix protrudes (or flowers) into the vagina of the mare.

The lining of the cervix consists of a series of folds or crypts, as shown in Fig. 1.12. These crypts are continuous with the smaller folds in the uterine endometrium and enable the significant expansion of the cervix required at parturition (Ginther, 1992; Kainer, 1993).

1.6. The Uterus

The uterus of the mare is a hollow muscular organ joining the cervix and the Fallopian tubes (Fig. 1.12). This upper part of the tract including the uterus is attached to the lumbar region of the mare by two broad ligaments, outfoldings of the peritoneum, on either side of the vertebral column. The broad ligaments provide the major support for the reproductive tract (Fig. 1.13) and can be divided into

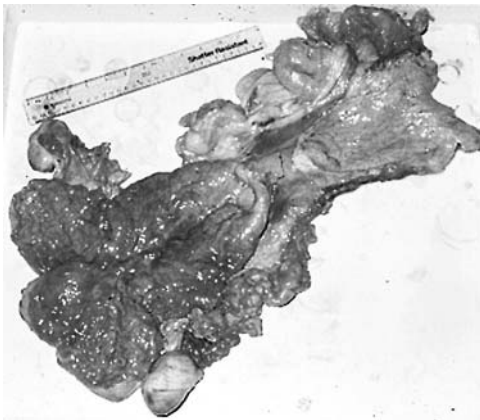


Fig. 1.12. The internal surface of the uterus, illustrating the endometrial folds of the uterine lining.

three areas: the mesometrium, attached to the uterus; the mesosalpinx, attached to the Fallopian tubes; and the mesovarium, attached to the ovaries (Ginther, 1992).

The uterus is divided into two areas, the body and the horns. The body of the uterus normally measures 18–20 cm long and 8–12 cm in diameter and divides into two uterine horns, which are approximately 25 cm long and which reduce in diameter from 4–6 cm to 1–2 cm as they approach the Fallopian tubes (Fig. 1.14). The size of the uterus is affected by age and parity, older multiparous mares tending to have larger uteri. The uterus of the mare is termed a simplex uterus bipartitus, due to the relatively large size of the uterine body compared with the uterine horns (60:40 split). This differs from that in other farm livestock, where the uterine horns are the more predominant feature. The lack of a septum dividing the uterine body is also notable (Hafez and Hafez, 2000). *In situ* the uterine walls are flaccid and intermingle with the intestine, the only lumina present being those formed between the endometrial folds.

The uterine wall (Fig. 1.13) consists of three layers: an outer serous layer (perimetrium) continuous with the broad ligaments, a central muscular layer (myometrium) and an inner mucous-membrane lining (endometrium). The central myometrial layer consists of external longitudinal muscle fibres, a central vascular layer and internal circular muscle fibres. It is this central myometrial layer that allows the considerable expansion of the uterus during pregnancy and provides the force for parturition. The inner endometrium is arranged in 12–15 longitudinal (Fig. 1.12) folds and

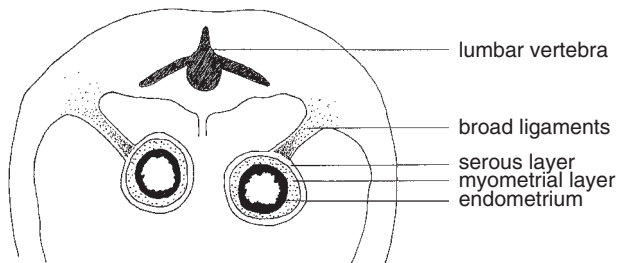


Fig. 1.13. Cross-section through the abdomen of the mare illustrating the reproductive-tract support provided by the broad ligaments.

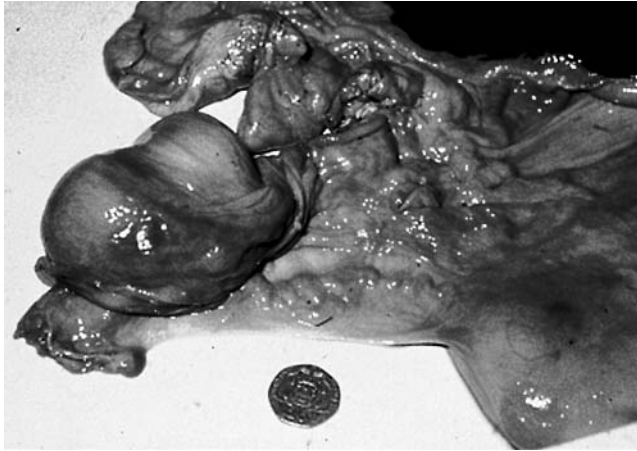


Fig. 1.14. The convoluted Fallopian tube running through the mesovarian section of the broad ligaments, from the uterine horn on the right to the ovary on the left.

comprises luminal epithelial cells, stroma of connective tissue or lamina propria and associated epithelial glands and ducts. Collagenous connective-tissue cores support these folds. The activity and therefore appearance of these endometrial glands are dependent on the cyclical hormonal changes. It is this endometrial layer that is largely responsible for supporting the developing conceptus and for placental attachment and development (Ashdown and Done, 1987; Ginther, 1992, 1995; Kainer, 1993; Sertich, 1998).

1.7. The Utero-tubular Junction

The utero-tubular junction is a constriction or sphincter formed by a high concentration of muscle cells from the circular myometrium of the Fallopian tube. The junction, which appears as a papilla in the endometrium, separates the end of the uterine horns from the beginning of the Fallopian tubes (Fig. 1.15). Fertilization takes place in the Fallopian tubes, and only fertilized ova can pass through this junction and on to the



Fig. 1.15. The utero-tubular junction in the mare, as seen from the uterine horn side (the dark colour of the uterine endometrium is not natural but allows easier identification of the utero-tubular junction).

uterus for implantation and further development. Fertilized ova appear to actively control their own passage, possibly via a localized secretion of prostaglandin E (PGE) (Ball and Brinsko, 1992), leaving the unfertilized ova on the Fallopian-tube side of the junction. These then gradually degenerate (Ginther, 1992; Kainer, 1993; Fig. 1.14).

1.8. The Fallopian Tubes

The mare has two Fallopian tubes or oviducts of 25–30 cm in length, which are continuous with the uterine horns (Fig. 1.14). The diameter of these tubes varies slightly along their length, being 2–5 mm at the isthmus end, nearest the uterine horn, and increasing to 5–10 mm at the ampulla, nearest the ovary. The division of the Fallopian tube between the isthmus and ampulla is approximately equal. The Fallopian tubes lie within peritoneal folds, which form the mesosalpinx part of the broad ligaments. They have walls very similar in structure to the uterus, but thinner, composed of three layers: the outer, fibrous, serous layer, continuous with the mesosalpinx; a central myometrial layer of circular and longitudinal muscles fibres; and an inner mucous membrane. Fertilization takes place in the ampulla, a region lined with fimbriae (hair-like projections), which act to waft unfertilized ova into the ampulla to await the sperm and to waft fertilized ova out of the ampulla and on towards the utero-tubular junction. The ampulla of each Fallopian tube ends in the infundibulum, a funnel-like opening close to the ovary (Sisson, 1975).

The infundibulum in the mare is closely associated with a specific part of the ovary termed the ovulation fossa, which is unique to the mare and is the only site of ova release; in other mammals ovulation may occur over the whole surface of the ovary. The infundibulum is, therefore, relatively hard to distinguish in the mare, not being so evident as a funnel-shaped structure surrounding the whole ovary. The infundibulum is lined, like the ampulla, by fimbriae, which attract and catch the ova, guiding them towards the entrance of the Fallopian tubes (Ginther, 1992; Kainer, 1993).

1.9. The Ovaries

The ovaries of the mare are both cytogenic and endocrine in function, producing gametes (ova) and hormones. They are evident as two bean-shaped structures situated ventrally to (below) the fourth and fifth lumbar vertebrae and supported by the mesovarium part of the broad ligaments. They make the total length of the reproductive tract in the mare in the region of 50–60 cm. In the sexually inactive stage, i.e. during the non-breeding season, the mare's ovaries measure 2–4 cm in length and 2–3 cm in width and are hard to the touch, due to the absence of developing follicles. During the sexually active stage when the mare is in season, they increase in size to 6–8 cm in length and 3–4 cm in width; they are also softer to the touch, due to the development of fluid-filled follicles (Fig. 1.16). Older, multiparous mares tend to show larger ovaries up to 10 cm in length.

The convex outer surface or border of the ovary is attached to the mesovarian section of the broad ligaments (Fig. 1.16) and is the entry point for blood and nerve supply; the concave inner surface is free from attachment and is the location of the ova fossa. The whole ovary is contained within a thick protective layer, the tunica albuginea, except for the ova fossa. The tissue of the ovary in the mare is arranged as the inner cortex (active gamete-producing tissue) and the outer medulla (supporting tissue). Ova release at ovulation occurs only through the ova fossa, and all follicular and corpora lutea development occurs internally, within the cortex of the ovary (Witherspoon, 1975). The mare differs in these aspects from other mammals, in which the medulla and cortex are reversed, ovulation occurring over the surface of the ovary and all follicular and corpora lutea development occurring on the outer borders. Rectal palpation, as a clinical aid to assess reproductive function in the mare, is not, therefore, as easy to perform as it is in other farm livestock, for example, the cow. However, with the advent of ultrasound, reproductive assessment of ovarian characteristics in the mare is now quite accurate (Ginther, 1992, 1995; Kainer, 1993; Sertich, 1998; Hafez and Hafez, 2000).



Fig. 1.16. The ovaries of the mare. Note the difference in size between the ovary on the left, which is inactive, and the one on the right, which is active. The concave surface (position of the ova fossa) and the convex surface (entry point for blood and nerve supply) of the ovary are clearly seen.

1.9.1. Follicular development and ovulation

The ovary is made of two basic cell types: interstitial cells (stroma), which provide support; and germinal cells, which produce the ova. The number of potential ova contained within the female ovary is dictated prior to birth; subsequently no addition to the pool of ova can be made. These very immature ova are termed oogonia, and there are many more than an individual will use within her reproductive lifetime. These oogonia, with their full complement of chromosomes (64) and surrounded by a single layer of epithelial cells, are termed primordial follicles. At birth, the ovary contains many thousands of these primordial follicles. After birth and prior to puberty, some oogonia start development to primary oocytes, and these, surrounded by their epithelial, or granulosa, cells, undergo the first stages of meiosis. They then await puberty, when hormone secretion from the anterior pituitary drives their further development.

From puberty onwards, primary oocytes develop and complete the final stages of meiosis at varying rates, designed to ensure that a regular supply of developed follicles

is available for ovulation every 21 days during the breeding season. Not all primary follicles are destined to ovulate, as many are wasted along the way, degenerating and becoming atretic; normally (in 75% of cases) just one reaches the stage ready for ovulation (Davies Morel and O'Sullivan, 2001).

As the primary follicle develops, its surrounding epithelial cells differentiate into follicular epithelial cells, which secrete follicular fluid, filling the cavity surrounding the oocyte. The follicle grows in size as fluid accumulation increases. The primary oocyte now also increases in size and develops a thick, outer, jelly-like layer, the zona pellucida; it is now termed a secondary oocyte and has a haploid number of chromosomes (32). The secondary oocyte now becomes associated with one inner edge of the follicle and lies on a mound of follicular cells called cumulus oophorus. The stroma immediately surrounding the follicle becomes organized into a double-lined membrane, the theca membrane, the inner layer of which is vascularized, the outer layer not. The follicles continue to develop (normally to in excess of 3 cm in diameter) and are termed Graafian follicles (Fig. 1.17).

This seems to be a critical stage in equine follicular development, for now a

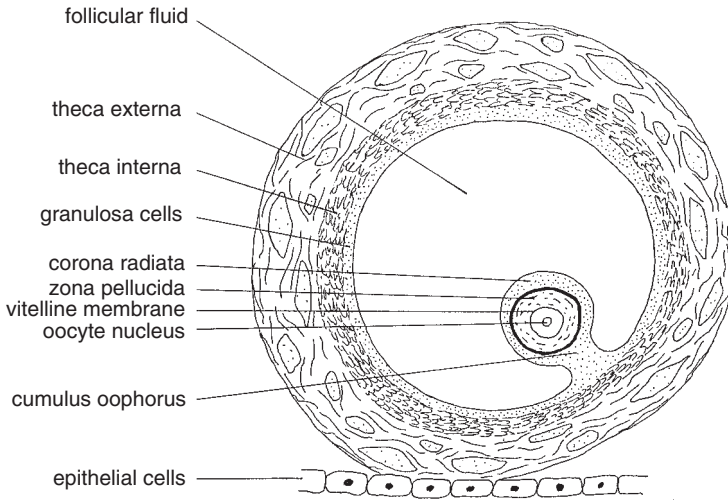


Fig. 1.17. The equine Graafian follicle.

decision is made as to which of the developing follicles is destined to develop further for ovulation and which ones will degenerate. In 75% of cases only one follicle is destined to ovulate and hence develop further. In 23% of cases two follicles may develop further and ovulate, the remainder being triplets or more (Davies Morel and O'Sullivan, 2001). This decision seems to be governed by the follicle's ability to respond to elevated levels of circulating oestrogen, follicle-stimulating hormone (FSH) and luteinizing hormone (LH), though the exact mechanisms are unclear (Fay and Douglas, 1987; Roy and Greenwald, 1987).

In those follicles destined to ovulate, follicular diameter increases and at the same time they appear to move within the stroma of the ovary and orientate themselves to await ovulation through the ova fossa. Ovulation of the mature follicle occurs in two stages, which normally (99% of occasions) occur concurrently. The two stages are follicular collapse and ova release. The whole process may take from a matter of seconds up to a few hours, with ova release occurring at the later stages of ovulation (J. Newcombe, Wales, 2001, personal communication). The ova and follicular fluid are

released through the ova fossa to be caught by the infundibulum and passed down the Fallopian tube for potential fertilization. Ovulation of follicles of diameter less than or greater than 3 cm does occur but this is the exception (Sirosis *et al.*, 1989; Ginther and Bergfeldt, 1993).

After the release of the ova and follicular fluid, the old follicle collapses and the theca membrane and remaining follicular epithelial cells become folded into the old follicular cavity. Bleeding, from the theca interna, occurs into the centre of this cavity, forming a clot. This clot, the theca cells and any remaining follicular epithelial cells make up the corpus luteum (CL) or yellow body. Blood capillaries and fibroblasts then invade the CL. It is initially a reddish-purple colour. As the CL ages, it becomes browner in colour and, if the mare is not pregnant, regresses to yellow and then white (corpus albicans) as it becomes non-functional. The luteal tissue is then gradually replaced with scar tissue (Fig. 1.18; Vogelsang *et al.*, 1987; Del Campo *et al.*, 1990; Kainer, 1993; Pierson, 1993; Ginther, 1995; Sertich, 1998; Hafez and Hafez, 2000). Figures 1.19–1.21 show sections through a mare's ovary, illustrating the presence of developing follicles and CL.

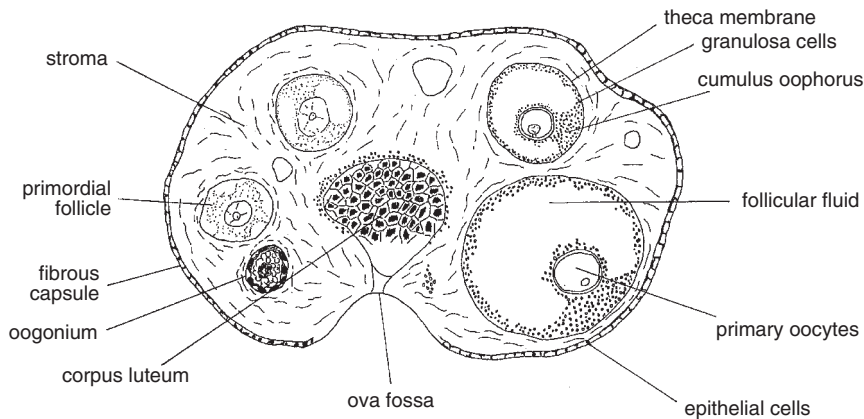


Fig. 1.18. Diagrammatic representation of follicular development and ovulation within the ovary.



Fig. 1.19. Cross section taken through the two ovaries pictured in Fig. 1.16. Note in the active ovary the corpora lutea (dark mass at the top left) and the large follicle (hollow or space at the top right).

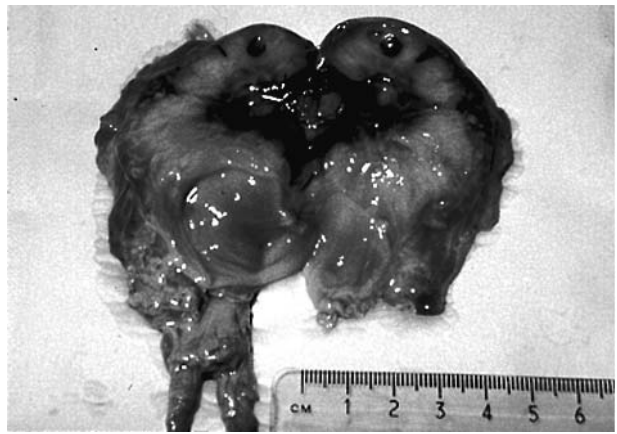


Fig. 1.20. A cross section taken through an active ovary illustrating the presence of a large follicle (3 cm in diameter) at the bottom of the ovary.



Fig. 1.21. A cross section taken through an active ovary illustrating a large corpus luteum at the top of the ovary.

1.10. Conclusion

It can be concluded that the reproductive tract of the mare is a remarkable system, designed

not only to maximize the chance of fertilization and subsequent maintenance of the resulting conceptus in a sterile environment, but also to expel that conceptus successfully at term.

2

The Reproductive Anatomy of the Stallion

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2.1. Introduction

This chapter details the anatomy and function of the stallion reproductive system. Further, more detailed, accounts may be found in other texts, such as Ashdown and Done (1987), Sack (1991), Ginther (1995), Dyce *et al.* (1996), Samper (1997), Bone (1998), Turner (1998), Davies Morel (1999), Chenier (2000) and Hafez and Hafez (2000). Figure 2.1 illustrates the main structures of the stallion's reproductive tract. Figure 2.2 shows the reproductive system of the stallion after slaughter and Fig. 2.3 provides a diagrammatic representation of this plate. These organs will be discussed individually below.

2.2. The Penis

The penis of the stallion may be divided into the glans penis, the body or shaft and the roots. In the resting position it lies retracted and hence protected within its sheath, or prepuce, out of sight; it is held in this position by muscles. The prepuce is a double-folded covering to the penis, which folds back on itself to give two folds of protection. Within the inner fold lies the end of the penis, the glans penis (or rose), giving this sensitive area additional protection. Protruding, by 5 mm, from the centre of the glans penis, lies the exit of the urethra. Around this protrusion lies the urethral fossa and, below it, a dorsal

Fig. 2.1. Diagrammatic representation of the reproductive system of the stallion.

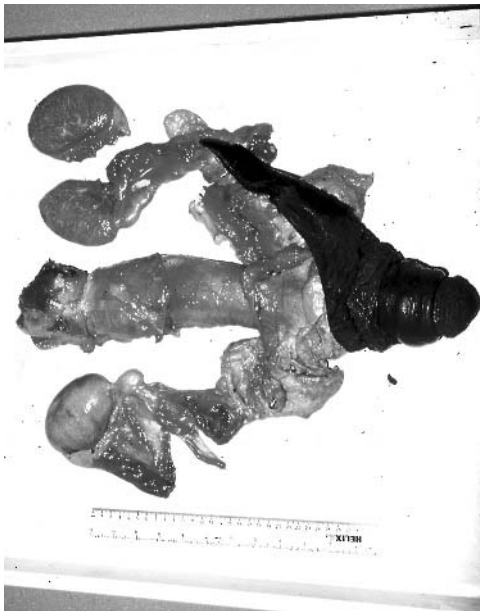


Fig. 2.2. The stallion's reproductive tract after slaughter and dissection, the accessory glands are not included (see also Fig. 2.3).

diverticulum, both of which are often filled with smegma, a red-brown secretion of the prepubital glands lining the prepuce plus epithelial debris. These areas also provide an ideal environment for bacteria, often harbouring venereal disease (VD) bacteria, such as *Klebsiella pneumoniae*, *Taylorella equigenitalis* and *Pseudomonas aeruginosa*.

The penis of the stallion is attached to the lower part of the pelvic bone by two roots. Here at its origin the penis is held in position by a ligament attachment to the pelvis. The urethra, running from the bladder, connects with the vas deferens and runs between the two roots before entering the body of the penis. The body of the stallion penis contains a large percentage of erectile, as opposed to fibrous, tissue and as such it is termed haemodynamic (reacts to increasing blood pressure). Figure 2.4 illustrates a cross-section view through the body area of the stallion penis.

Figure 2.4 shows that the main body of the penis is divided into two sections, the lower corpus cavernosus urethra and the upper

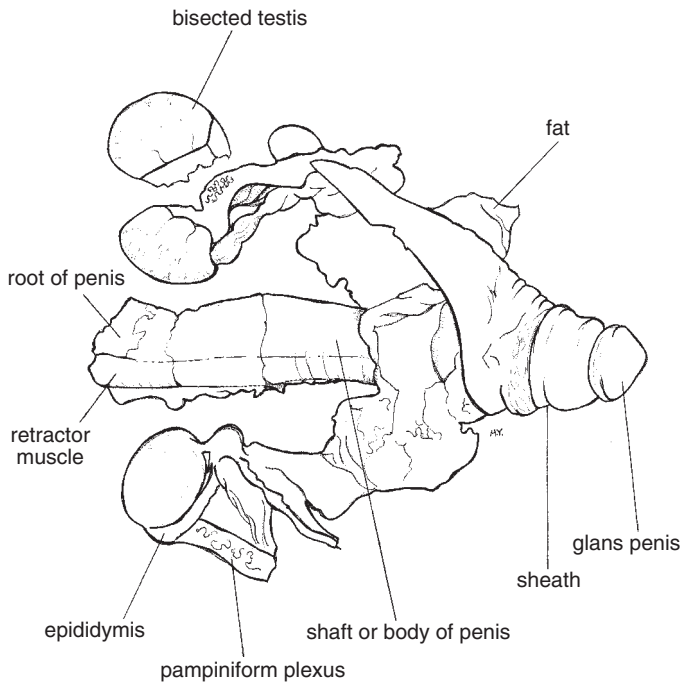


Fig. 2.3. The stallion's reproductive tract: a diagrammatic representation of Fig. 2.2.

corpus cavernosus penis. Through the corpus cavernosus urethra runs the urethra, surrounded by some trabeculae (sheets of connective tissue) enclosing small areas of erectile tissue, all enclosed within the bulbospongiosus muscle. The corpus cavernosus penis, which is the largest part of the penis, contains a dense network of trabeculae, associated muscle tissue and scattered cavities, making up the major erectile tissue of the penis. The corpus cavernosus penis is contained within the tunica albuginea, a fibro-elastic capsule or sheet, which maintains the integrity of the penis but still allows the doubling in size seen at erection. Finally, running along the bottom of the penis is a retractor muscle, contraction of which returns the penis to within the prepuce. The major erectile tissue of the glans penis is a continuation of the corpus cavernosus urethra, the corona glandis (Fig. 2.4b). This large amount of erectile tissue, which is not confined by the fibro-elastic capsule of the corpus cavernosus penis, allows the greater expansion (up to three times) of this area at ejaculation.

2.2.1. Erection, ejaculation and emission

During the process of erection, the first reaction is a relaxation of the penile muscles and retractor muscle, allowing the penis to extrude from its sheath. Blood engorgement of the erectile tissue of the penis results in an initial turgid pressure, followed by a drawing up of the penis against the pelvis reducing the exit of venous blood. This, plus an increase in general blood pressure, further increases penile blood pressure to one of intromission pressure. It is essential that the penis is at intromission pressure before entering the mare; if not, it may cause permanent damage to the stallion or at least reduce his enthusiasm for future covering. The sequence of events leading to erection is the result of central nervous system stimulation, in response to the presence of a mare in oestrus or other sexual stimulation.

The culmination of erection, ejaculation, is the result of contraction of the muscle walls of the epididymis, vas deferens and the accessory glands, passing sperm plus semi-

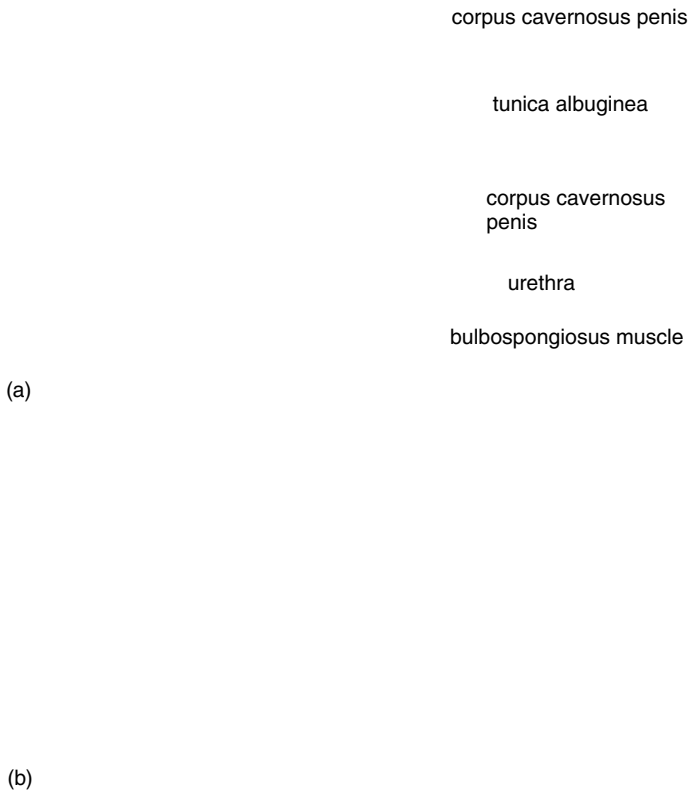


Fig. 2.4. Cross-section through the penis of the stallion: (a) the main body of the penis; (b) the glans penis.

nal fluid to the penis. Exit of semen from the penis is termed emission and is achieved by contraction of the muscle fibres within the penis, as well as contractions of the vas deferens. Emission is in the form of six to nine jets of semen, the initial three jets containing the majority of the biochemical components, forming the sperm-rich part of the ejaculate. In addition to the sperm-rich fraction, pre-sperm and post-sperm fractions are evident, both of which are smaller in volume and do not contain viable sperm (Kosiniak, 1975).

At full erection, the penis doubles in size to 80–90 cm in length and 10 cm in width. At ejaculation, the glans penis triples in size, helping to open the cervix and so allow

sperm deposition into the uterus and possibly prevent initial leakage of semen from the mare. The glans penis has to return to near-normal size before the stallion is able to leave the mare. Failure to allow time for the glans penis to return to normal can cause damage to the mare and/or stallion. Problems may be encountered in overzealous stallions that demonstrate enlargement of the glans penis prior to entry into the mare; in such cases intromission is not safe until the glans penis has returned to its normal size. Such problems are especially evident in stallions of high libido, particularly if they have only a limited work load (Ginther, 1995; Turner, 1998; Davies Morel, 1999).

2.3. The Accessory Glands

The accessory glands are a series of four glands (some authors consider there to be three glands, excluding the ampulla) situated between the end of the vas deferens and the roots of the penis. Collectively these glands are responsible for the secretion of seminal plasma.

2.3.1. Seminal plasma

Seminal plasma is the major fluid fraction of semen. Seminal plasma provides the substrate for conveying the sperm to the mare and ensuring final maturation. Some of its major functions are the provision of energy and protection from changes in osmotic pressure and from oxidization. It also contains a gel, which forms a partial clot in semen, the function of which is unclear (Davies Morel, 1999).

Males of most species have the same series of accessory glands, the relative size of the glands reflecting the relative importance of their secretions within the seminal plasma.

2.3.2. The bulbourethral glands

The bulbourethral glands, or Cowper's glands, are the accessory glands situated nearest to the roots of the penis. They are paired and oval in structure, approximately 2 cm by 3 cm and lying either side of the urethra. Their secretions are clear, thin and watery and form part of the main sperm-rich fraction; they may also contribute to the pre-sperm fraction. As part of the pre-sperm fraction, they aid in clearing urine and bacteria collected within the urethra prior to ejaculation. Their secretion may also act as a lubricant, easing the passage of sperm (Mann, 1975; Weber and Woods, 1993).

2.3.3. The prostate

The prostate gland is a bilobed structure with a single exit to the urethra, situated between the bulbourethral glands and the

ampulla. Prostate secretions in the stallion are alkaline and high in proteins, citric acid and zinc. The significance of these is unclear. Secretions of the prostate gland make a significant contribution to the pre-sperm fraction (Little and Woods, 1987).

2.3.4. The vesicular glands

The vesicular glands, or seminal vesicles, are again paired in structure and lie either side of the bladder, about 16–20 cm in length. They are lobed and can be compared to large walnuts in external appearance. They secrete a major amount of seminal plasma, with a high concentration of potassium, citric acid and gel. Their secretions form part of both the sperm-rich and the gel-like post-sperm fractions. Their function and therefore the volume of secretion are dependent upon circulating testosterone concentrations. As such, their contribution to seminal plasma significantly declines during the non-breeding season (Thompson *et al.*, 1980; Weber *et al.*, 1990).

2.3.5. The ampulla

The ampulla glands are paired outfoldings of the vas deferens situated where it meets the urethra. The secretion of the ampulla glands contains a high concentration of ergothionine, an antioxidantizing agent, protecting susceptible chemicals in semen from oxidization. The secretions of the ampulla are largely associated with the pre-sperm fraction.

2.3.6. The vas deferens

The vas deferens connects the epididymis of the testis to the urethra before passing the accessory glands and on into the penis. It has a diameter of 0.5–1 cm, with a thick muscular wall, made up of three layers of muscle; an inner oblique, middle circular and outer longitudinal (Fig. 2.5). These muscle layers actively propel the sperm plus surrounding fluid from the testis to the penis. The lumen of the duct is small and lined with folds,

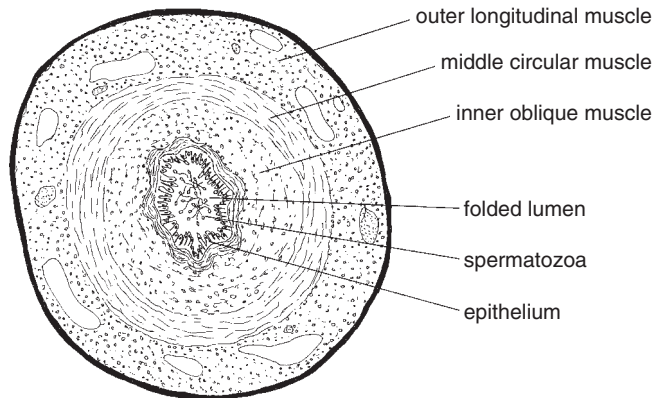


Fig. 2.5. Cross-section through the vas deferens of the stallion.

especially near the epididymis, maximizing the surface area and so aiding sperm storage and the reabsorption of testicular fluids.

The vas deferens, the testicular nerve supply, arterial and venous blood-vessels and the cremaster muscles pass out of the body cavity through the inguinal canal (Fig. 2.6). The cremaster muscle, which is divided into internal and external sections, is responsible for drawing the testis up towards the body in response to cold, fear, etc.

2.4. The Epididymis

The epididymis in the stallion lies over the top of the testis, as illustrated in Fig. 2.3 and Fig. 2.7. It consists of long convoluted tubules and is subdivided into three sections: the head (caput); the body (corpus); and the tail (cauda). The head of the epididymis is connected by several ducts to the rete testis; as it continues towards the tail end, these ducts merge and form a single duct, the vas

Fig. 2.6. Cross-section through the inguinal connection between the testis and the stallion's body.

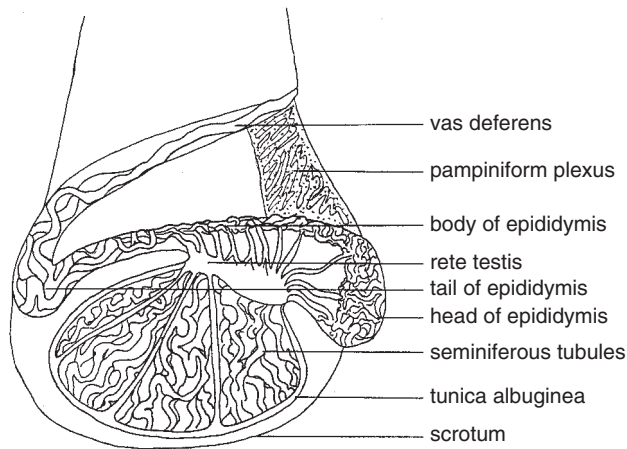


Fig. 2.7. Diagrammatic illustration of a vertical cross-section through the testis of the stallion.

deferens. The lining of these tubules is highly folded, very similar to that of the epididymal end of the vas deferens. These folds have additional microvilli, increasing the surface area still further and so facilitating the reabsorption of testicular secretions in order to concentrate the sperm and increase its storage capacity (Thompson, 1992).

It is essential that all sperm spend a period of time, up to 7 days, within the epididymis in order to mature. Such maturation is essential so that released sperm are capable of further development (capacitation) within the female tract, enabling them to fertilize the awaiting ova. If they are not passed up to the vas deferens as a result of ejaculation, they degenerate and are reabsorbed, allowing a continual supply of fresh sperm in order to maintain fertilization rates (Thompson *et al.*, 1979). The tail end of the vas deferens, therefore, acts as a storage site for sperm and also contributes towards seminal plasma by secreting glycerylphosphorylcholine (GPC) (Samper, 1995a).

2.5. The Testis

The testes are gametogenic (the site of sperm production) and endocrine in function. Figure 2.2 and Figs 2.7 and 2.8 illustrate the structure of the testis.

The testes hang outside the body of the stallion in order to maintain a temperature of approximately 3°C below that of body temperature (i.e. 35–36°C rather than 39°C). Sperm production is maximized at this lower temperature. Increases in testicular temperature due to disease or inflammation of the scrotum, testis or epididymis result in a significant decrease in spermatogenesis. This is transitory but the duration of testicular dysfunction is related to the duration of



Fig. 2.8. The dissected stallion testis illustrating testicular tissue, the epididymis, vas deferens and pampiniform plexus.

temperature elevation. The testis temperature is normally controlled by means of the cremaster muscles, which can retract the testis up towards the body, along with an abundance of scrotal sweat glands and the arteriovenous counter-current heat-exchange mechanism provided by the pampiniform plexus (Roberts, 1986b; Friedman *et al.*, 1991). The pampiniform plexus allows the testicular artery to divide into a dense capillary network before it enters the testis (Fig. 2.3, Figs 2.7 and 2.8). As such, it comes into close contact with the returning venous supply, also divided up into a capillary network. Blood entering the testis, therefore, loses its heat to the venous return. Such an arrangement ensures that the testicular artery cools down prior to entry into the testis and the testicular vein is warmed up prior to its re-entry into the main body.

The testes lie within a skin covering, termed the scrotum, under which lies the tunica vaginalis, which is continuous with the peritoneal lining of the body cavity up through the inguinal canal. In the fetus, the testes descend from a position near the kidney, through the inguinal canal and into the scrotum at or soon after birth (Fig. 2.9). The failure of one or both testes to descend fully may not necessarily mean a stallion is infertile, as the testis within the body cavity may still produce sperm; sperm count,

however, is normally much reduced. A stallion with only one descended testis is termed a cryptorchid or rig. The condition may be further defined as unilateral, bilateral, inguinal or abdominal, depending on whether one or both testes have failed to descend and how far descent has progressed (Figs 19.2 and 19.3).

The testes of the stallion normally lie with the long axis horizontal unless drawn up towards the body, when they may turn slightly. The long axis is normally 6–12 cm with the height and width being 4–7 cm and 5–6 cm, respectively. Testes may weigh 300–350 g per pair. Their size increases allometrically with general body growth until final body size has been reached, at approximately 5 years of age.

Under the tunica vaginalis, a fibrous capsule, the tunica albuginea, surrounds each separate testis. Sheets of this fibrous tissue invade the body of the testis and divide it up into lobes. Each lobe is a mass of convoluted seminiferous tubules with intertubular areas (Figs 2.7 and 2.10). Each area is largely responsible for one of two functions, cytogenic (seminiferous tubules and Sertoli cells) or endocrine (intertubular tissue and Leydig cells). The Sertoli cells lining the seminiferous tubules act as nurse cells, nourishing and aiding the developing spermatozoa in the lumen of the tubules. In addition, they are also phagocytic, digesting

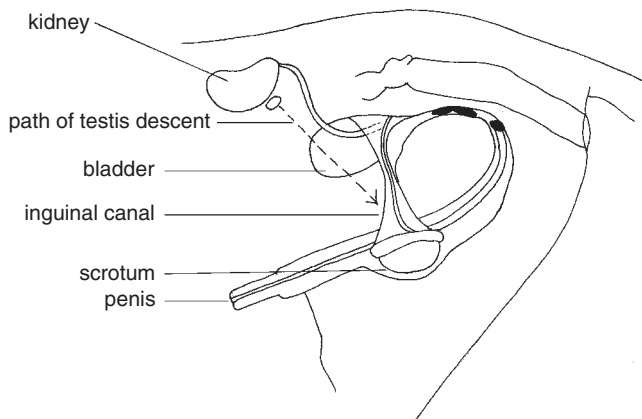


Fig. 2.9. The normal passage of descent of the testis in the stallion.

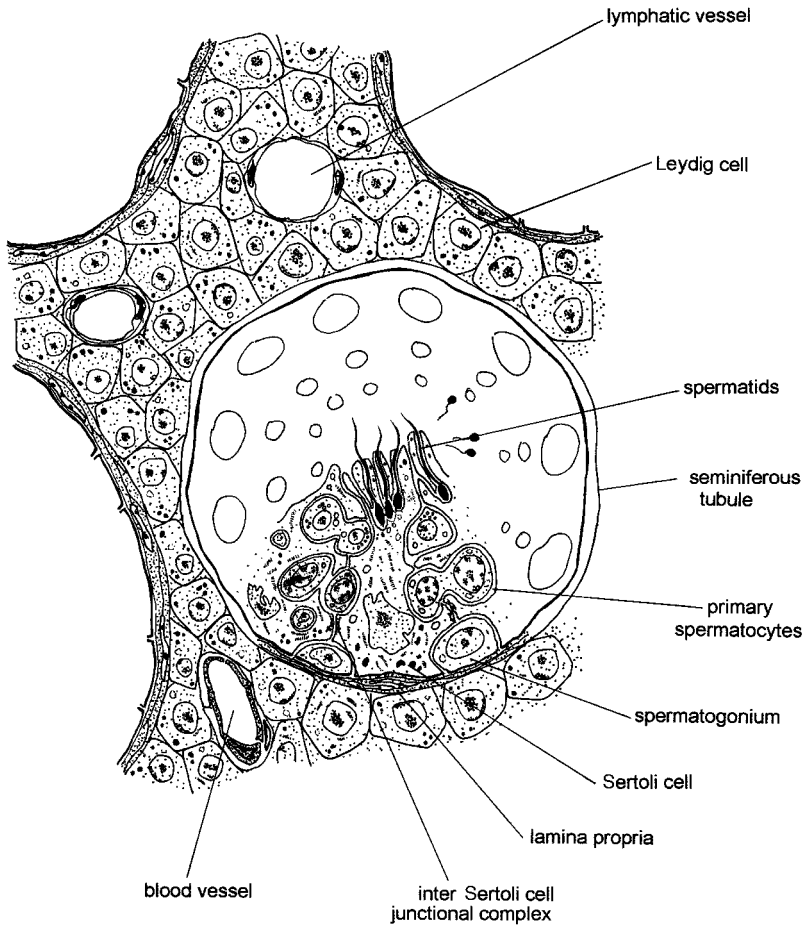


Fig. 2.10. A cross-section view through a seminiferous tubule within the stallion's testis, illustrating the gradual meiotic division of spermatogonia to spermatozoa.

degenerating germ cells and residual bodies; they secrete luminal fluid and proteins, and also form a blood–testis barrier, providing protection for the sperm from immunological rejection and cell-to-cell communication. The number of Sertoli cells varies with season, being significantly greater during the breeding season, when they are actively involved in sperm production. As such, Sertoli cells have a dominant control over sperm production (Johnson and Tatum, 1989). This increase in Sertoli-cell number is accompanied by a corresponding increase in seminiferous-tubule length (Johnson and Nguyen, 1986).

The Leydig cells, found in the intertubular spaces around the seminiferous tubules, secrete hormones that are largely responsible for sperm production and the development of general male bodily characteristics and behaviour (Setchell, 1991; Amann, 1993a,b).

2.6. Sperm

As discussed, sperm are produced within the seminiferous tubules and are supported or nursed by Sertoli cells. They start as underdeveloped germ cells or spermatogonia attached to the wall of the seminiferous tubules, and

then progressively develop into primary spermatocytes, secondary spermatocytes, spermatids and finally mature sperm (Amann, 1981b; Amann and Graham, 1993; Fig. 2.10). As they develop, they migrate, within their Sertoli cells, away from the wall of the seminiferous tubules towards the open lumen. Once mature, they are then freed by their Sertoli cell, released into the lumen and pushed along the seminiferous tubules by rhythmic contractions of the tubules and the surrounding secretory fluid. By this stage they have lost a considerable amount of cytoplasm but have developed tails. The tails are not functional until epididymal maturation has occurred. The whole cycle of development takes 57 days and occurs in waves, ensuring a continual supply of mature sperm for ejaculation, providing the stallion is not overused. The mean daily sperm production of a mature stallion is in the order of $7-8 \times 10^9$ sperm and it takes 8–11 days for sperm to pass from the testis to the exterior (Dinger and Noiles, 1986b; Johnson *et al.*, 1997; Davies Morel, 1999). Total sperm production is related to testis size or volume, which can be assessed by using callipers or ultrasonically. The use of callipers in the stallion is not as easy as in other livestock, which have more pendulous testes. Ultrasonic assessment is preferred and gives a more accurate result (Love *et al.*, 1991).

Structurally, sperm consist of three areas: the head; the midpiece; and the tail (Fig. 2.11), with three distinct functions. The head is mainly made up of nuclear material, containing the haploid number of chromosomes (half the normal number, 32, to allow fusion with the ova to give the normal diploid complement of 64). The head of the sperm has a double membrane, the outer cell membrane and the inner nuclear membrane, except in the acrosome region at the top of the head, where there is an additional acrosome membrane. The importance of this membrane will become apparent when fertilization is considered, as it is responsible for the breakdown of the cell membrane and the nuclear membrane at fertilization, allowing the fusion of the male and female nuclei. The mid-piece of the sperm contains a high proportion of mitochondria, organelles within the cell that pro-

duce energy. The mid-piece is often termed the power plant of the sperm, providing the energy to drive the tail. The tail is made up of a series of muscle fibrils, equivalent to those found in the major muscle blocks of the body. Using the energy provided by the mid-piece, the tail is whipped from side to side, producing a wave like motion (Amann and Graham, 1993; Davies Morel, 1999).

2.7. Semen

Semen is the term applied to seminal plasma plus sperm, and in the stallion is a milky-white gelatinous fluid. The sperm concentration of semen varies with the fraction examined. As previously mentioned, there are three identifiable fractions, the pre-sperm, sperm-rich and post-sperm fractions. The pre-sperm fraction is the initial fraction and contains no sperm. Its function is to clean and lubricate the urethra prior to ejaculation. Stale urine and bacteria readily collect in the urethra. The high concentration of bacteria in this fraction means that its collection should be avoided when collecting semen for artificial insemination.

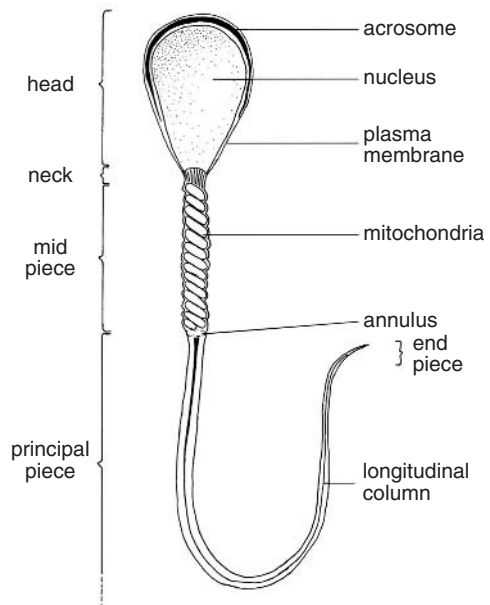


Fig. 2.11. A typical stallion sperm.

The sperm-rich fraction is the major deposit by the stallion and commences as soon as the glans penis swells to force entry into the cervix. This fraction is normally 40–80 ml in volume and contains 80–90% of the sperm and the biochemical components of semen.

The third fraction is the post-sperm or gel fraction. Its volume varies enormously from none at all to 80 ml and is dependent on libido: the higher the libido, the greater the gel fraction. Season also affects the gel-fraction volume, this being lower in the non-

breeding season. Breed and previous use also have an effect. If a stallion is used more than once per day, the second ejaculate frequently has half the gel fraction of the first. The volume of the gel-free fraction is also affected by age, as are sperm numbers and concentration, as illustrated in Table 2.1 (Squires *et al.*, 1979b).

At ejaculation, the stallion secretes semen in a series of up to nine jets, the average volume of semen and the sperm concentration decreasing with successive jets (Fig. 2.12; Tischner *et al.*, 1974; Kosiniak, 1975; Mann, 1975).

Table 2.1. The effect of age on the seminal characteristics of stallions. Means in the same row with different subscripts differ significantly. NS, not significant. (From Squires *et al.*, 1979b.)

Characteristics	Age (years)			P
	2–3 (n = 7)	4–6 (n = 16)	9–16 (n = 21)	
Seminal volume (ml)				
Gel	2.1	5.1	13.3	NS
Gel-free	14.2 _a	26.2 _b	29.8 _b	0.05
Total	16.2 _a	31.4 _{ab}	43.2	< 0.01
Spermatozoa				
Concentration (10 ⁶ ml ⁻¹)	120.4	160.9	161.3	NS
Total (10 ⁹)	1.8 _a	3.6 _{ab}	4.5 _b	< 0.05
Motility (%)	55.0	63.1	59.9	NS
pH	7.68 _a	7.64 _{ab}	7.59 _b	< 0.05

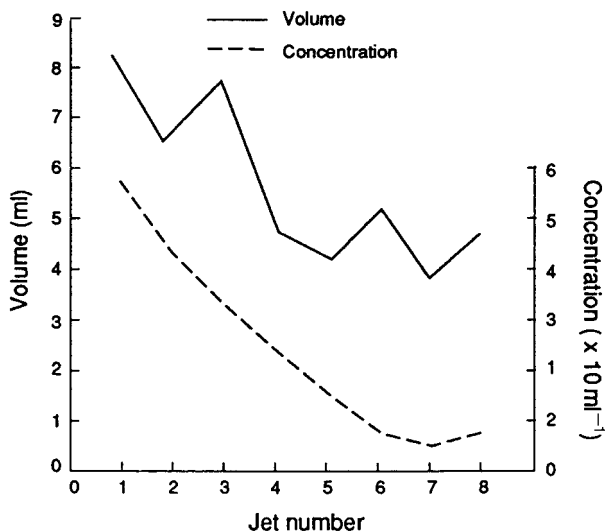


Fig. 2.12. Volume and sperm concentration of successive jets (from Kosiniak, 1975).

2.8. Conclusion

It can be concluded that the male reproductive tract is specifically designed for the efficient production, storage and subsequent

deposition of sperm within the female tract. It also ensures that the sperm are deposited in a medium (seminal plasma) that is able to provide all the elements for their survival, so maximizing the chances of fertilization.

3

Endocrine Control of Reproduction in the Mare

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3.1. Introduction

The mare is naturally a seasonal breeder, showing sexual activity only during the spring, summer and autumn months. This is termed the breeding season, and the non-breeding season is termed anoestrus. On average, the breeding season lasts from April until November in the northern hemisphere and October to May in the southern hemisphere (eight to 12 cycles). However, the breed of the mare has a significant effect, as has individual variation. Ponies and the large heavier breeds, especially of the cold-

blooded type, tend to show shorter seasons than the finer, more hot-blooded types, e.g. the Thoroughbred and lighter riding horses. It is not unknown for well-fed stabled horses to be still showing regular oestrous cycles as late as the January prior to going into seasonal anoestrus. During her breeding season, the mare shows a series of spontaneous oestrous cycles at regular intervals and is therefore termed a seasonal polyoestrous spontaneous ovulator. The rest of her breeding cycle is summarized in Fig. 3.1, which illustrates the major milestones in the mare's reproductive life.

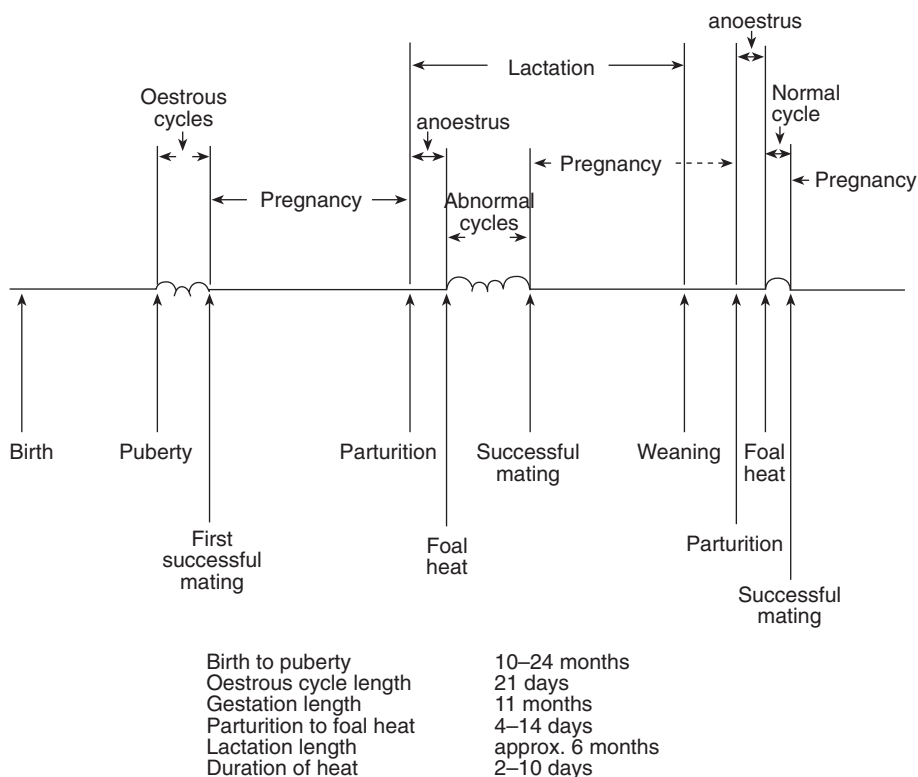


Fig. 3.1. A summary of the breeding cycle of the mare.

The oestrous cycles of the mare commence at puberty (10–24 months of age). Each cycle lasts, on average, 21 days (range 20–22 days). Each cycle is a pattern of physiological and behavioural events under hormonal control and is divided into two periods: oestrus, when the mare is sexually receptive, normally 4–5 days; and dioestrus, when she will reject sexual advances, normally 16 days. On either side of truly receptive oestrus; two other phases have been suggested: pro-oestrus, as the mare comes into oestrus; and metoestrus, as the mare goes out. These periods are more evident in mares than in other farm livestock, as oestrus is longer and less distinct (Gordon, 1997). The exact times of these periods vary considerably between individuals and with season and age, tending to be longer in the transition periods into and out of the breed-

ing season and in older mares. In general, any variation in cycle length is due to a variation in the pro-oestrus, oestrus and metoestrus phases, rather than in dioestrus. For example, a cycle length of 20 days is likely to be due to 15 days' dioestrus and 5 days' pro-oestrus, oestrus and metoestrus; for a mare showing a 26-day cycle, the respective times would be 15 days and 11 days (J. Newcombe, Wales, 2001, personal communication). Ovulation normally occurs 24–36 h before the end of oestrus and is denoted by day 0. Days 1–21 then denote the remainder of the cycle until ovulation recurs (Ginther, 1992). The cycle may also be divided into luteal (corpus luteum (CL) is dominant) and follicular (follicle development is dominant) phases.

Oestrous cycles continue throughout the mare's lifetime and only cease during the

non-breeding season. The mare is an efficient breeder, showing oestrous cycles during lactation, unlike some other seasonal breeders, such as the ewe, and she is, therefore, capable of being pregnant and lactating at the same time. The mare shows her first oestrus after foaling often within 4–10 days; this oestrus is termed her foal heat. After the foal heat, the mare may start to show her regular 21-day cycles, but in many cases, due to the effects of lactation, it takes a while for the system to settle down to a regular pattern again (Mathews *et al.*, 1967; Allen, 1978; Ginther, 1992; Watson *et al.*, 1994b).

For ease of understanding, the following discussion on the oestrous cycle of the mare has been divided into two sections: the associated physiological changes; and the behavioural changes.

3.2. Physiological Changes

The major physiological events associated with reproductive activity in the mare are

endocrine changes, which in turn govern and drive the other physiological changes, as well as her behavioural activity.

3.2.1. The endocrinological control of the oestrous cycle

The endocrinological control of the oestrous cycle is governed by the hypothalamic–pituitary–gonad axis; a similar axis controls the reproductive activity of the stallion. The gonads, in the case of the mare, are the ovaries (Fig. 3.2). Overriding the whole of this control mechanism is the effect of photoperiod; decreasing day length causing oestrous cycles to cease and increasing day length causing them to occur. The plane of nutrition and environmental temperature are also thought to play a part. Day length is perceived by the pineal gland in the base of the brain, which, by means of the hormone melatonin, controls the activity of the hypothalamic–pituitary–ovarian axis. Melatonin is produced nocturnally by the pineal gland

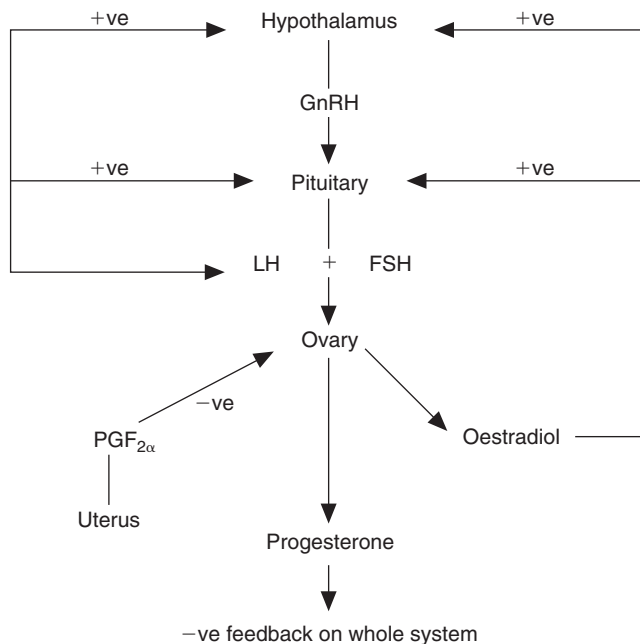


Fig. 3.2. The hypothalamic–pituitary–ovarian axis, which governs reproduction in the mare. GnRH, gonadotrophin-releasing hormone; LH, luteinizing hormone; FSH, follicle-stimulating hormone; PGF_{2α}, prostaglandin F_{2α}.

and, under the influence of short day lengths, dominates the reproductive system, inhibiting the activity of the axis. As day length increases, inhibition of the axis is removed, allowing gonadotrophin-releasing hormone (GnRH) to be produced by the hypothalamus, so driving luteinizing hormone (LH) and follicle-stimulating hormone (FSH) production by the pituitary (Fitzgerald *et al.*, 1987). LH is released in a pulsatile manner and the frequency of these pulses is seen to increase from 0.38 to 4.74 pulses day⁻¹ as the mare moves from anoestrus to the first ovulation of the season (Fitzgerald *et al.*, 1985). Melatonin is secreted by the pineal gland in two phases: photophase (daytime); and scotophase (night-time). It therefore demonstrates a circadian secretion, with the highest levels of secretion being evident during the scotophase. The presence or absence of daylight is perceived by the pineal gland via neural messages from the retina of the eye. In the absence of light, the conversion of tryptophan to melatonin is driven (Grubaugh, 1982; Kilmer *et al.*, 1982; Cleaver *et al.*, 1991; Sharp and Cleaver, 1993). The exact means by which melatonin controls the hypothalamus is unclear, but it seems likely to involve dopamine and endogenous opioids, including β -endorphin (Kilmer *et al.*, 1982; Aurich *et al.*, 1994, 1995; Guerin and Wang, 1994; Besognek *et al.*, 1995).

Prolactin, another major seasonally affected hormone, seems to be responsible in the horse for changes in metabolic rate, increasing the efficiency of food conversion during the winter months, a time of food deprivation. Especially evident in the more native breeds, this demonstrates an innate ability of the equine body to anticipate environmental conditions and respond accordingly (Argo and Smith, 1983; Morley *et al.*, 1983; Evans *et al.*, 1991). Prolactin is thought, therefore, to translate primarily the changes in day length to seasonal changes in non-reproductive physiology, with only a limited effect of reproductive seasonality. The link or mechanism by which melatonin and prolactin secretions interact is better known in other seasonal breeders than in horses (Johnson, 1986a, 1987b; Thompson *et al.*,

1986; Roser *et al.*, 1987; Thompson and Johnson, 1987; Worthy *et al.*, 1987; Evans *et al.*, 1991; Nequin *et al.*, 1993; Besognek *et al.*, 1995).

When day length is appropriate, the hypothalamus is driven to produce GnRH. GnRH release, in common with other reproductive hormones, is tonic and pulsatile in manner. Tonic secretion relates to the background continual level of secretion, whereas pulsatile secretion is superimposed upon this as a series of pulses or episodes of higher levels. Both the level of tonic secretion and the amplitude and frequency of episodes can vary throughout the cycle. An increase in episode amplitude, frequency or tonic secretions causes an increase in average hormone concentrations. Eighty per cent of GnRH released is passed directly down a specialized portal system, the hypothalamic-pituitary portal vessels, to have a direct effect on the anterior pituitary (adenohypophysis), with 20% passing back to the central nervous system to affect behaviour. The level of GnRH in the mare's circulatory system is, therefore, relatively low, as its passage to the anterior pituitary is directed along these specialized portal vessels. In response to GnRH, the anterior pituitary produces the gonadotrophins, FSH and LH, the target organ for which is the ovaries (Alexander and Irvine, 1993; Irvine and Alexander, 1993a,b).

3.2.1.1. Follicle-stimulating hormone

FSH, as its name suggests, is responsible for the stimulation of follicle development. It is passed into the general circulatory system of the mare and its concentration suggests a biphasic release, with elevated levels on days 9–12 of the cycle and at ovulation. The largest rise occurs around ovulation and starts at day 15 with levels of 4 ng ml⁻¹, increasing to reach concentrations of 9 ng ml⁻¹ during oestrus (Fig. 3.3). This biphasic release adds weight to the theory that follicular development in the mare occurs over a 21-day period, in contrast to that in the sheep, cow and pig, where it is much shorter (3–6 days). It is likely that the peak at ovulation may serve two purposes: (i) to complete final follicle development prior to ovulation;

and (ii) to start the development of a new cohort of follicles in readiness for the next ovulation in 21 days (Fay and Douglas, 1987; Alexander and Irvine, 1993; Bergfeldt and Ginther, 1993; Ginther and Bergfeldt, 1993; Irvine and Alexander, 1993a, 1994). Up to ten follicles may be initially affected by the rise in FSH, but only a select one or two develop to a stage that can react to the final message to ovulate.

Figure 3.3, along with the subsequent graphs illustrating plasma hormone concentrations, are drawn to give an appreciation of the relative, rather than absolute, hormone concentrations. As with GnRH, all the hormones discussed here in relation to reproduction are secreted in a tonic and episodic fashion. The following series of graphs only indicate the average hormone concentrations. Absolute levels reported vary considerably between different scientific reports. Where known, concentrations are discussed within the text.

3.2.1.2. Inhibin and activin

It is suggested that the subsequent decline in FSH is brought about, at least in part, by the secretion of inhibin by large follicles as

they near ovulation. Inhibin acts as a negative feedback on FSH production by modulating the anterior pituitary response to GnRH, in the form of reducing FSH secretion. Activin has also been isolated in follicular fluid and is reported to have a similar but positive feedback effect, again specifically on FSH secretion (Piquette *et al.*, 1990; Nett, 1993b).

3.2.1.3. Oestrogen

As the follicles develop, they secrete oestrogens, the major one being oestradiol-17 β , an ovarian steroidal oestrogen, produced from cholesterol by an interrelationship between the theca and the granulosa cells within the developing follicle. The theca cells convert cholesterol to progesterone, which diffuses across to the neighbouring granulosa cells, where it is converted to oestradiol-17 β . This final conversion within the granulosa cells depends upon the enzyme aromatase, whose activity is FSH-dependent. Oestradiol-17 β is secreted into the main circulatory system and 24–48 h prior to ovulation reaches a peak of 10–15 pg ml⁻¹, dropping to basal levels immediately post-oestrus. This decline in oestrogen secretion

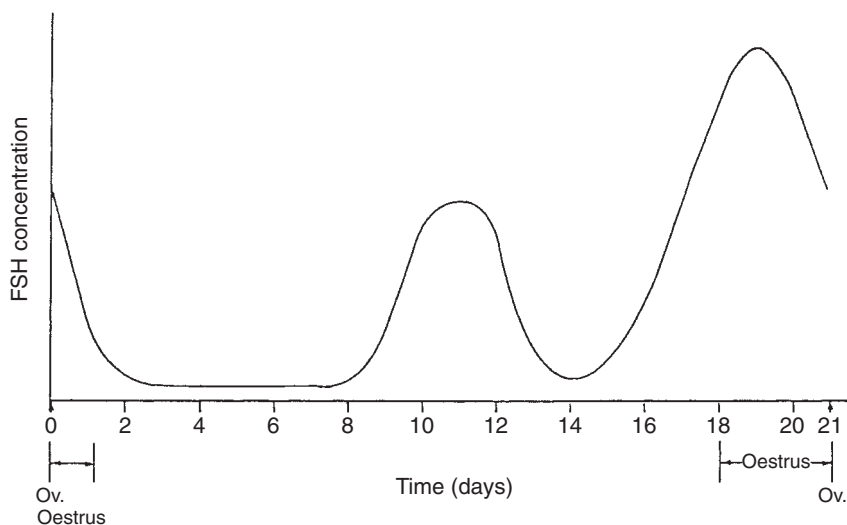


Fig. 3.3. Variations in the relative plasma concentrations of follicle-stimulating hormone (FSH) in the non-pregnant mare.

is associated with the release of the granulosa cells into the follicular fluid as part of the ovulation process, leaving the theca cells to produce progesterone but no granulosa cells for its conversion to oestradiol-17 β (Tucker *et al.*, 1991). Oestrogens are responsible for the behavioural changes in the mare associated with oestrus and sexual receptivity.

As FSH levels rise, oestradiol levels also increase, both reaching a peak within oestrus, thus ensuring that maximum follicular development in readiness for ovulation is synchronized with oestrus (Knudsen and Velle, 1961; Garcia *et al.*, 1979; Nett, 1993a; Weedman *et al.*, 1993; Fig. 3.4).

3.2.1.4. Luteinizing hormone

The message to ovulate is LH, which, like FSH, is secreted by the anterior pituitary. At oestrus, both tonic and episodic concentrations of LH rise to a peak. However, it is the increase in episodic pulse frequency and amplitude that is largely responsible for peak LH concentrations. Receptors to LH on the follicular theca cells increase in number

as LH concentrations rise. Increasing LH thus drives additional androgen-precursor production, providing more progesterone to diffuse across to the granulosa cells for conversion to oestradiol-17 β , which in turn further drives oestrous behaviour. Hence, rising LH levels induce increasing oestradiol-17 β secretion, further ensuring the synchronization of ovulation and oestrous behaviour. LH levels begin to rise from their basal levels of less than 1 ng ml⁻¹, with a pulse frequency of 1.4 pulses 24 h⁻¹, several days before the onset of oestrus. They then reportedly reach a peak of 10–16 ng ml⁻¹ just after ovulation (Whitmore *et al.*, 1973). It has been reported by some that LH not only drives final follicular development and induces ovulation but is also involved in the formation of the CL, possibly explaining why peak concentrations are not reached until just after ovulation. LH declines from peak concentrations to low dioestrous levels within a few days of ovulation (Pattison *et al.*, 1972; Evans *et al.*, 1979; Pantke *et al.*, 1991; Alexander and Irvine, 1982, 1993; Irvine and Alexander, 1993a, 1994; Aurich *et al.*, 1994; Fig. 3.5).

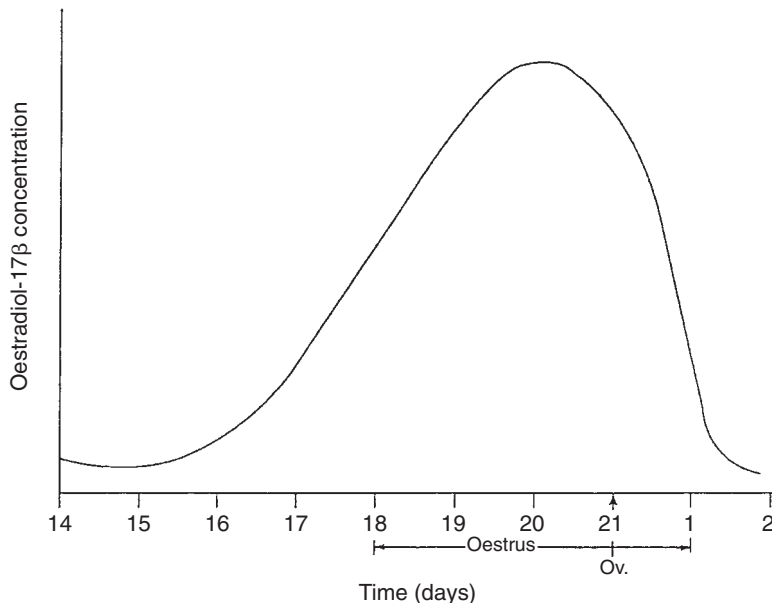


Fig. 3.4. Variations in the relative plasma concentrations of oestradiol in the non-pregnant mare.

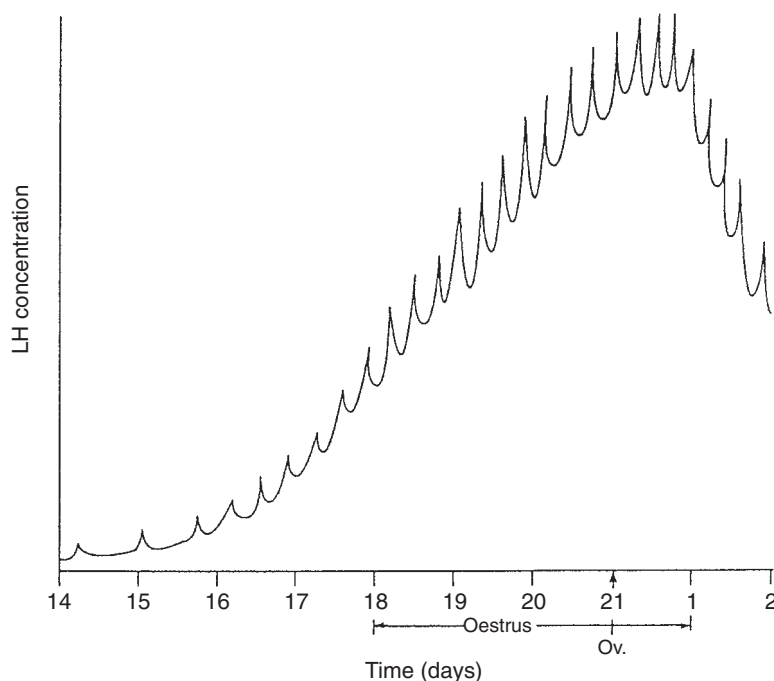


Fig. 3.5. Variations in the relative plasma concentration of luteinizing hormone (LH) in the non-pregnant mare.

3.2.1.5. Progesterone

Ovulation of a follicle results in the formation of a CL within the collapsed follicle lumen left after the ovum (or ova) and follicular fluid have been released. The luteal tissue contained within the CL is largely derived from the old theca cells and, as such, secretes progesterone. Progesterone levels, therefore, rise postovulation, commencing within 24–48 h. Maximum concentrations (10 ng ml^{-1}) are reached 5–6 days postovulation and are maintained until day 15–16 of the oestrous cycle. If the mare has not conceived, progesterone levels drop dramatically 4–5 days prior to the next ovulation to give basal levels again during oestrus (Fig. 3.6).

Progesterone has an inhibitory effect on the release of gonadotrophins in most farm livestock. Oestrus cannot begin, therefore, until progesterone levels have fallen to below 1 ng ml^{-1} . However, the block to gonadotrophin release in the mare is not so complete. Elevated progesterone levels appear to have

an inhibitory effect on the release of LH, preventing any rise in LH until progesterone levels decline. However, progesterone does not seem to have such an inhibitory effect on FSH. Indeed, unique to the mare, a second rise of FSH is apparent 10–12 days after ovulation, despite elevated progesterone concentrations.

If the mare fails to conceive, progesterone levels must decline in order to allow the mare to return to oestrus and ovulate on day 21. In order to induce the decline in progesterone, a message has to be received by the reproductive system of the mare informing her that there is no conceptus present. The messenger is the hormone prostaglandin $F_{2\alpha}$ ($\text{PGF}_{2\alpha}$).

3.2.1.6. Prostaglandin $F_{2\alpha}$

$\text{PGF}_{2\alpha}$ is difficult to measure in the peripheral circulatory system because of its short half-life and pulsatile manner of release. However, $\text{PGF}_{2\alpha}$ has a metabolic breakdown product, prostaglandin F metabolite (PGFM), which has a longer half-life and so is easier

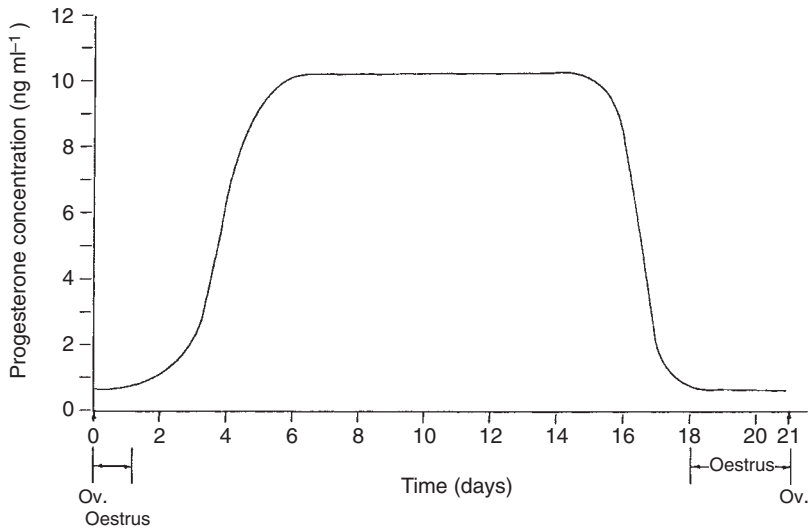


Fig. 3.6. Variations in the relative plasma concentrations of progesterone in the non-pregnant mare.

to measure in blood serum and plasma. As such, it closely mimics changes in $\text{PGF}_{2\alpha}$. Using levels of PGFM as a guide, it can be seen that $\text{PGF}_{2\alpha}$ levels rise between days 14 and 17 postovulation, immediately before progesterone levels start to decline. In mares

suffering from retained CL or those that are pregnant, no such rise is detected. $\text{PGF}_{2\alpha}$ is known to be secreted by the uterine endometrium and causes luteolysis (destruction) of the CL, thus causing progesterone levels to decline (Fig. 3.7). In the mare, $\text{PGF}_{2\alpha}$

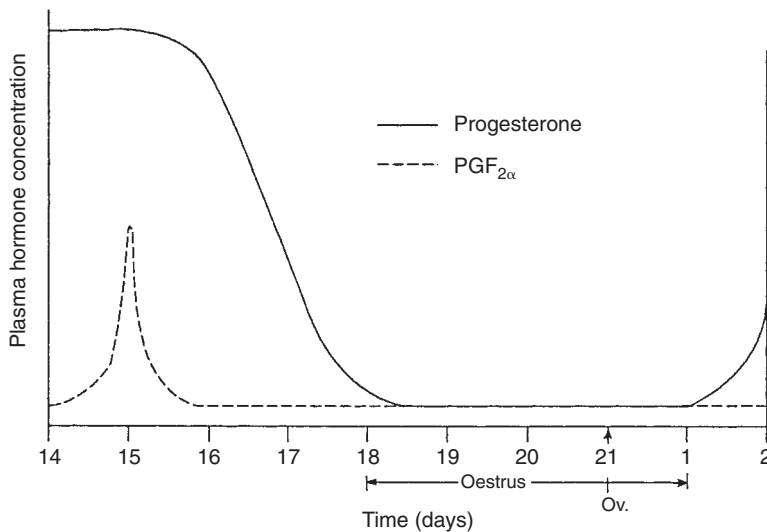


Fig. 3.7. Variations in the relative plasma concentrations of prostaglandin $\text{F}_{2\alpha}$ ($\text{PGF}_{2\alpha}$) and progesterone in the non-pregnant mare.

reaches the ovary via the main circulatory system, not by a local counter-current transport system as seen in the ewe and cow. This can be demonstrated by hysterectomy, as, in the case of the mare, removal of the uterine horn ipsilateral to (on the same side as) the CL does not result in maintenance of that CL (Ginther and First, 1971).

The decline in progesterone levels, in response to $\text{PGF}_{2\alpha}$ secretion, removes any inhibition of gonadotrophin release, allowing the hormone changes associated with oestrus and ovulation to commence.

3.2.1.7. Oxytocin

The message of non-pregnancy is also thought to involve the release of oxytocin. Oxytocin in this instance may be produced by the CL and is transported via the main circulatory system to the uterus, seemingly enhancing the release of $\text{PGF}_{2\alpha}$ (Tetzke *et al.*, 1987; Nett, 1993b).

3.2.1.8. Summary

Figure 3.8 summarizes simply the major fluctuations in hormone concentrations

during a single oestrous cycle of a non-pregnant mare.

The following is a summary of the major events that occur in the mare's oestrous cycle.

Day 0	Ovulation LH rising FSH falling Oestradiol falling Oestrus
Day 1	LH peak Metaoestrus
Day 2	Oestrus ends Dioestrus begins LH declining FSH approaching basal levels Oestradiol approaching basal levels
Day 5	Progesterone rising
Day 9	Progesterone at maximum
Day 11	FSH rising
Day 13	FSH peak
Day 15	FSH at basal levels $\text{PGF}_{2\alpha}$ peak Progesterone begins to fall
Day 16	FSH rising Progesterone falling

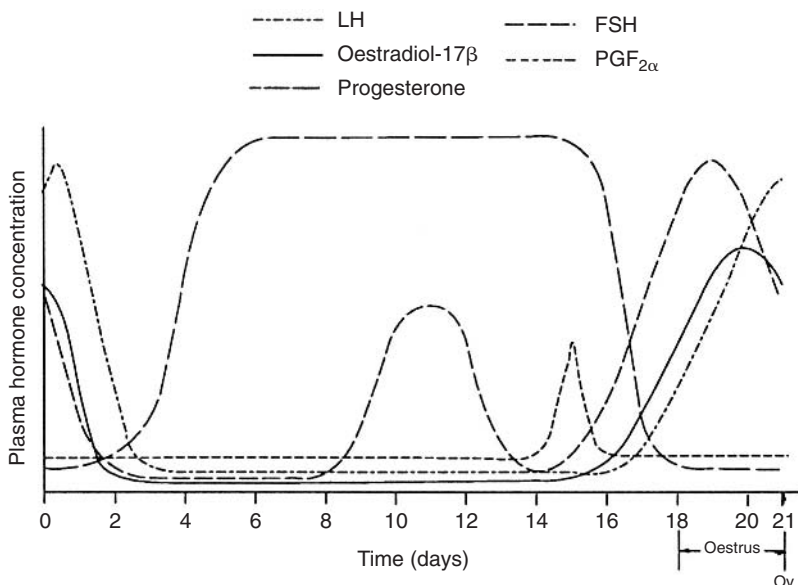


Fig. 3.8. A simplified summary of the major plasma hormone concentration changes during the oestrous cycle of the non-pregnant mare. LH, luteinizing hormone; FSH, follicle-stimulating hormone; $\text{PGF}_{2\alpha}$, prostaglandin $\text{F}_{2\alpha}$.

Day 18	FSH rising
	Progesterone basal
	Oestradiol rising
	Pro-oestrus
	LH rising
Day 20	Progesterone basal
	FSH peak
	LH rising
	Oestradiol reaching a peak
	Oestrus
Day 21/0	Ovulation
	LH rising
	FSH falling
	Oestradiol falling
	Oestrus

3.2.2. Physiological changes of the genital tract

As well as the cyclical changes in hormone concentration, changes in the mare's reproductive tract are also seen; these are driven by the fluctuations in hormone levels. In the uterus, the epithelium proliferates during early dioestrus in preparation for embryo implantation. The epithelial cells are activated and appear tall and columnar during dioestrus, becoming deactivated and cuboidal in nature as the next early oestrus approaches (Ginther, 1992). The epithelial glands also change configuration with the cycle, becoming more active and secretive and appear vacuolated during dioestrus. Characteristic changes in uterine secretions can be detected by ultrasonic scanning typically producing 'cartwheel'-like images at oestrus (Pycock, 2000; J. Newcombe, Wales, 2000, personal communication). Leucocyte concentrations within the uterus also vary, increasing during oestrus and so helping to combat infection at a vulnerable time. This increase is thought to be associated with elevated circulating oestradiol concentrations at this time.

Uterine myometrial contractility also varies with the cycle, being more active during oestrus. This activity encourages the expulsion of the uterine exudates and secretion at a time when the tract is most vulnerable to uterine infection.

In general, the changes within the uterus result in an increase in uterine wall thickness

and turgidity as the mare goes from oestrus into dioestrus in preparation for the imminent implantation of an embryo. If pregnancy does not occur, luteolysis results in a reduction of the thickness of the uterine wall and a reversal of these changes as oestrus approaches.

Cervical changes also occur within the cycle. Cervical appearance, as viewed by a vaginoscope, can be used as a diagnostic aid in the detection of reproductive activity. During dioestrus, the cervix is tightly closed, forming a tight seal against entry into the uterus. Its appearance is white, firm and dry. During oestrus, the cervix relaxes, opening the cervical seal to allow entry of the penis at mating. During oestrus, the cervix appears moist, red and dilated as the secretions of the uterine epithelial cells and cervical cells increase (Warszawsky *et al.*, 1972). The presence or absence of these secretions within the vagina is also indicative of the stage of the oestrous cycle. It is often very hard to insert a vaginoscope into the vagina of a dioestrous mare, due to the thick, sticky nature of the secretions.

3.2.3. Variations in cyclic changes

The mare is notorious for variations or abnormalities in her reproductive cycle. This is in contrast to other farm livestock, which have been specifically bred over time for their ability to reproduce rather than to perform athletically.

A wide variation in the length of oestrus is evident between mares, the extremes being between 1 and 50 days. In general, a variation can be seen with the time of year, longer and less distinct oestrous periods being evident during the beginning and end of the breeding season. Nutritional intake also causes variation in oestrous length. When nutrition is limited, oestrus tends to be longer and less distinct, making it less likely that the mare will conceive during such a non-ideal time. This effect of poor nutrition may be an additional signal to the mare, indirectly indicating seasonal and therefore day-length changes (Sharp, 1980; Daels and Hughes, 1993).

The length of dioestrus also varies between mares, with the extremes being 10 days to several months. This delay is normally due to one of three reasons: first, a silent ovulation – ovulation occurred but it was not accompanied by oestrus, giving the impression that the mare has been in dioestrus for a prolonged period of time; second, the existence of a persistent CL – a CL that has not reacted to PGF_{2α} or has not received enough PGF_{2α} to elicit a response; or third, inactive ovaries, usually associated with the transition into or out of the non-seasonal state or true anoestrus. Other variations in the cycle do occur – for example, ovulation in dioestrus. LH is normally released in a low episodic fashion (1–4 ng ml⁻¹) during dioestrus; occasionally, however, these episodes are large enough to cause ovulation mid-cycle, despite the high dioestrous progesterone levels (Vandeplasseche *et al.*, 1979b). This evidence of dioestrous rises in LH and the previously discussed second FSH peak mid-cycle indicates that, unlike many other species, progesterone does not serve to block completely gonadotrophin release in the mare. The converse, oestrus with no ovulation, has also been reported, normally in mares out of the breeding season (Hughes and Stabenfeldt, 1977; Daels and Hughes, 1993). Covering at the foal heat is often unsuccessful, as fertility rates are normally low. Additionally the oestrous cycles following this foal heat have an increased chance of being disturbed, often showing prolonged oestrus and dioestrus, until steady cyclicity is achieved (Loy, 1980; Blanchard and Varner, 1993a; Camillo *et al.*, 1997).

The causes of many of these variations can be attributed to managerial or environmental influences, e.g. nutrition, temperature, day length, etc. Occasionally, they are due to genetic faults, lactational effects or embryonic death.

3.2.4. Multiple ovulation

Multiple ovulations – the release of more than one ovum per oestrus – are increasingly common in mares. Release of the ova may occur all within oestrus (synchronous) or occur over time, including early dioestrus

(asynchronous). All such ovulations may be considered as multiple ovulations as they have the potential to be fertilized and yield viable embryos (Ginther and Bergfelt, 1988). However, the more distant over time the ovulations occur, the less likely the chance of fertilization and those occurring more than 3 days apart rarely result in multiple conceptuses. The reported incidence of multiple ovulations in mares is very variable, at 0.83% to 42.8% (Arthur and Allen, 1972; Warszawsky, *et al.*, 1972; Wesson and Ginther, 1981; Ginther *et al.*, 1982; Newcombe, 1995). The issue of multiple ovulation and multiple pregnancies presents many dilemmas and is of significant economic importance to horse breeders; this issue will be further considered in Chapter 5.

3.3. Behavioural Changes

Cyclical, hormonal changes govern the mare's behavioural patterns in association with oestrus and dioestrus, elevated oestradiol concentrations and the absence or presence of progesterone being major factors, stimulating behavioural centres of the brain. As considered previously, GnRH is also known to play a minor role in oestrous behaviour (Irvine and Alexander, 1993b). There are many variations between individuals in the extent and strength of behavioural changes (Munro *et al.*, 1979). Details of the signs of oestrus and their interpretation are given in Chapter 13. A summary of the major behavioural changes is given below.

Oestrus initiated by elevated oestradiol concentrations:

Signs of oestrus	Dolcility
	Urination stance
	Lengthening and eversion of the vulva
	Exposure of the clitoris (winking)
	Tail raised
	Urine bright yellow with a characteristic odour
	Acceptance of the stallion's advances

Dioestrus evident in the absence of oestra-
diol:

Signs of dioestrus Hostility
 Rejection of stallion's
 advances

3.4. Conclusion

The prime aim of all the control mechanisms for the female reproductive system is to synchronize the physiological and behavioural changes associated with oestrus, in order to synchronize mating with ovulation and so achieve fertilization, and subsequently synchronize embryo and uterine development.

4

Endocrine Control of Reproduction in the Stallion

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4.1. Introduction

The stallion, like the mare, is a seasonal breeder but tends to show a less distinct season and, unlike her, if given enough encouragement, is capable of breeding all the year round. However, season does have an effect upon seminal volume, sperm concentration in the gel-free fraction, sperm per ejaculate, the number of mounts per ejaculate and reaction time to the mare (Pickett *et al.*, 1970, 1975a; Pickett and Voss, 1972; Johnson, 1991b). Figures 4.1–4.5 demonstrate the effect of season on reproductive parameters. Sperm production, unlike ova production, is a continual process and is not governed by cyclical hormonal changes.

As with the mare, reproductive activity commences at puberty and continues for the rest of the stallion's lifetime, though there is

some suggestion that semen quality declines after 20 years of age. The exact timing of puberty is unclear and varies with breed and development. Various researchers have used histological changes within the testis, especially in association with the Leydig cells, to indicate the timing of puberty. Ages of 1–2.2 years have been suggested using such parameters (Cornwall, 1972; Naden *et al.*, 1990; Clay and Clay, 1992). However, other work suggests that the colt may be up to 5 years old before full adult reproductive ability is reached. The characteristics tested were testicular weights, daily sperm production, testosterone concentrations and Leydig and Sertoli cell number and volume (Johnson and Neaves, 1981; Thompson and Honey, 1984; Berndston and Jones, 1989). Stallions of 3 years of age have normally reached puberty and are spermatogenically active

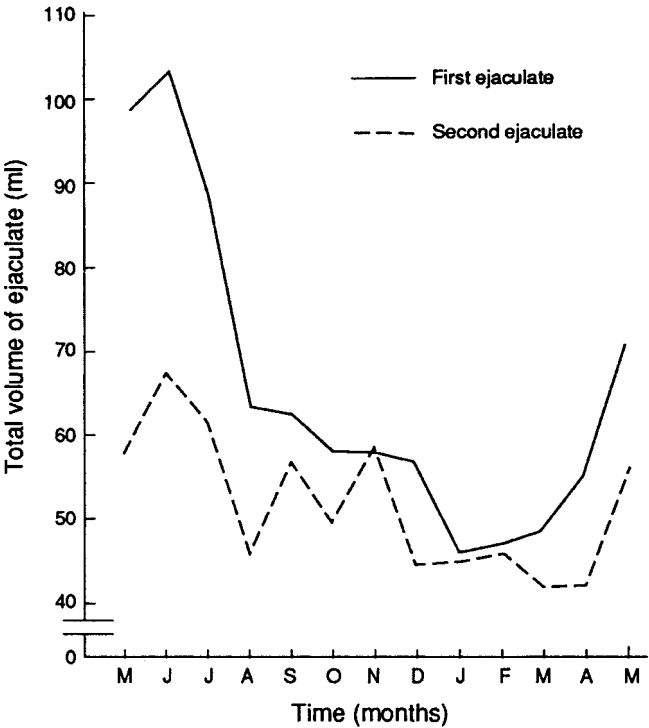


Fig. 4.1. Total seminal volume produced throughout the year (from Pickett and Voss, 1972).

Fig. 4.2. Number of sperm in gel-free semen fraction throughout the year (from Pickett and Voss, 1972).

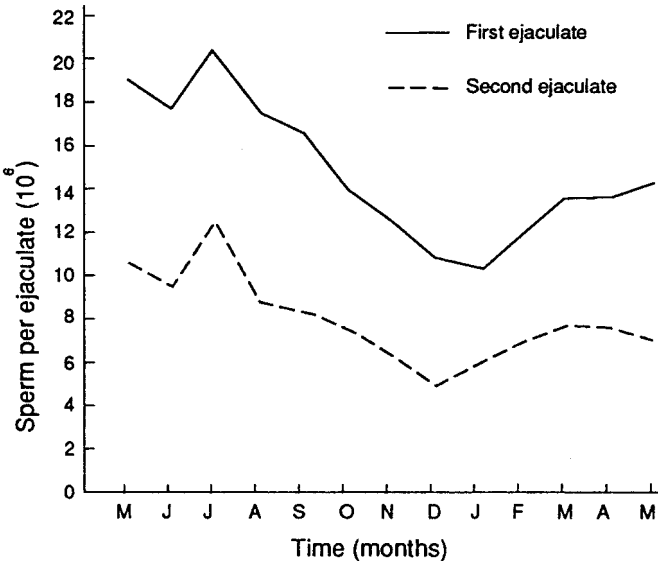


Fig. 4.3. Number of sperm per ejaculate throughout the year (from Pickett and Voss, 1972).

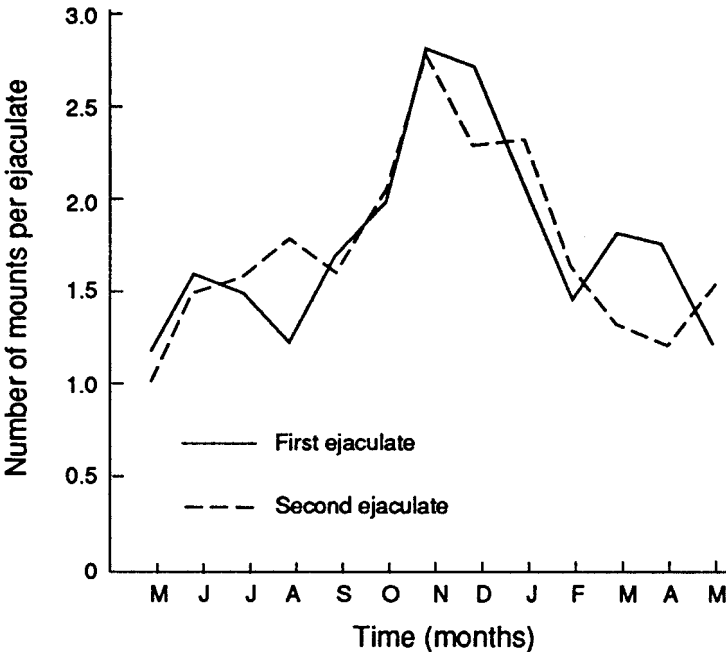


Fig. 4.4. Mean number of mounts required per ejaculate throughout the year (from Pickett and Voss, 1972).

and so perfectly capable of fertilizing mares covered; however, they have a limited sperm-producing capacity. By 5 years of age, they

are capable of producing adequate numbers of spermatozoa to cover a full complement of mares (Johnson *et al.*, 1991).

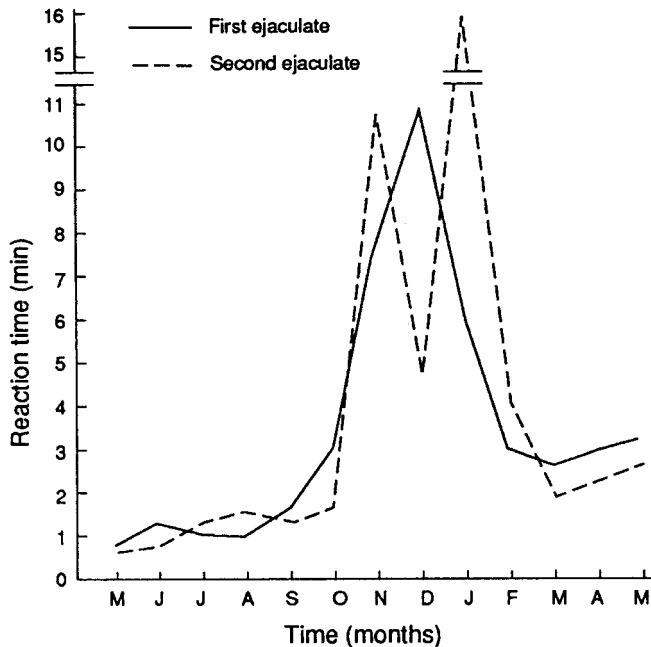


Fig. 4.5. The effect of season on sexual behaviour as measured by reaction time (from Pickett and Voss, 1972).

As with the mare, the reproductive activity of the stallion can be divided into physiological and behavioural changes and will be detailed in turn.

4.2. Physiological Changes

Hormone patterns are the major physiological changes associated with stallion reproductive activity and indeed govern the remaining physiological and behavioural changes.

4.2.1. The endocrinological control of stallion reproduction

Control of stallion reproduction is governed by the hypothalamic–pituitary–gonad axis, as seen in the mare, except in the stallion the testes are the gonads (Amann, 1981a). This axis is summarized in Fig. 4.6.

Environmental stimuli, in the form of day length and temperature, have an overriding effect on the above axis. Season is governed

by the secretion of melatonin from the pineal gland in response to day length, in a similar manner to that discussed for the mare (Thompson *et al.*, 1977; Burns *et al.*, 1982; Clay *et al.*, 1987). Both melatonin and photoperiod can be manipulated, as in the mare, to alter the timing of the breeding season, but overstimulation with artificial long days for a prolonged period of time results in refractoriness to the photo-stimulation (Clay *et al.*, 1987; Cox *et al.*, 1988; Argo *et al.*, 1991).

Again, as discussed in detail for the mare, prolactin concentrations are also affected by day length, increasing as day length increases ($4\text{--}5\text{ }\mu\text{g ml}^{-1}$ in summer, $1.7\text{ }\mu\text{g ml}^{-1}$ in winter). It is thought that prolactin is the means by which photoperiod controls coat growth and weight gain (Tucker and Wetterman, 1976).

4.2.1.1. Luteinizing hormone and follicle-stimulating hormone

The anterior pituitary is stimulated, via gonadotrophin-releasing hormone (GnRH), to produce follicle-stimulating hormone

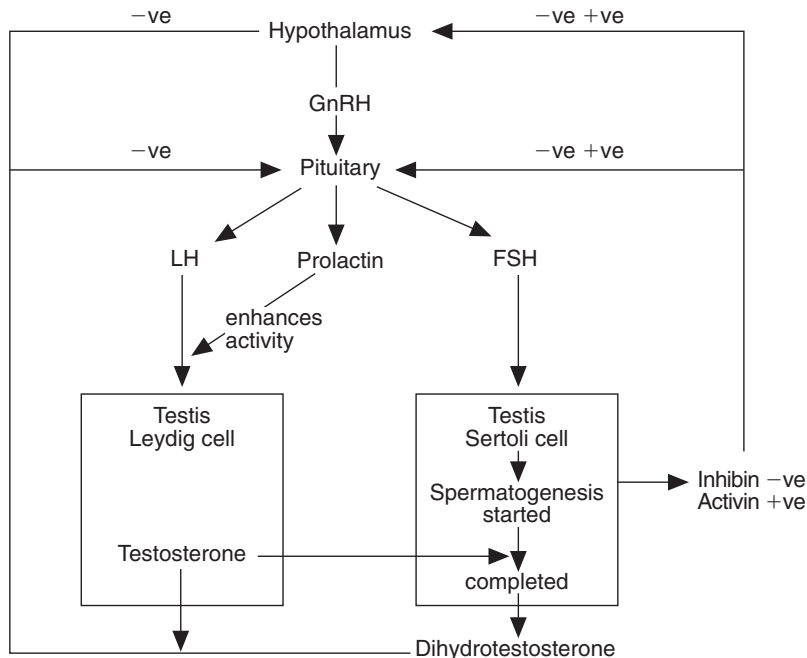


Fig. 4.6. The hypothalamic–pituitary–testis axis, which governs reproduction in the stallion. GnRH, gonadotrophin-releasing hormone; FSH, follicle-stimulating hormone.

(FSH) and luteinizing hormone (LH) (Irvine, 1984). The plasma concentrations of LH and FSH in the sexually active stallion are of the order of 3–4 ng ml⁻¹ and 7–7.5 ng ml⁻¹, respectively (Cupps, 1991; Seamens *et al.*, 1991; Amann, 1993b; Kainer, 1993).

4.2.1.2. Testosterone

The testes, which are the target organs for LH and FSH, consist of two major cell types, Leydig and Sertoli cells, plus associated structural tissue (Fig. 2.10). Leydig cells are found within the intertubular spaces or interstitial tissue of the testis and are responsible for the production of testosterone. These cells and, therefore, testosterone secretion are controlled by LH (Amann, 1981a). LH is produced in a pulsatile fashion, and so is testosterone. Such pulsatile release of testosterone means that a single blood sample for the hormone can give erroneous results; a hormone profile taken over a period of time and averaged is a much more accurate indication of true testosterone levels (Amann, 1993b).

Sertoli cells are found lining the seminiferous tubules and act as nurse cells to developing spermatids. Both FSH and testosterone drive these cells. FSH is known to start the process of spermatogenesis, developing spermatogonia to secondary spermatocytes. Testosterone then completes their development from secondary spermatocytes to spermatozoa, ready for passage to the epididymis for maturation (Davies Morel, 1999).

Additionally, testosterone controls the development of male genitalia, testis descent in the fetus or neonate, pubertal changes and accelerated growth, plus the maintenance and function of the accessory glands. It is also responsible for male libido and sexual behaviour by stimulation of the central nervous system, plus development of personality, stallion behaviour and muscular development. Testosterone also feeds back negatively on pituitary function to reduce the release of LH and FSH and hence its own production (Irvine *et al.*, 1986; Flink, 1988). A derivative of

testosterone, dihydrotestosterone, also feeds back negatively on the pituitary, as well as having a limited effect on the other testosterone-driven stallion characteristics. This negative feedback ensures that the system does not overrun itself. Testosterone may also be produced by a limited population of Leydig cells occasionally found in the wall of the vas deferens; this is evident in some geldings that have been successfully gelded but continue to demonstrate significant stallion characteristics. Testosterone is also produced by the adrenal glands in both the stallion and the gelding and it is this testosterone that is responsible for the continued, but reduced, male characteristics of the gelding.

Not only is the production of testosterone dependent upon season (Fig. 4.7), but also a diurnal rhythm is suggested. Testosterone concentrations have been reported to be elevated at 06.00 and 18.00 h. It has been postulated that in the wild this ensures that mating activity is greatest at dawn and dusk, times of least risk to stallions and mares from predators (Pickett *et al.*, 1989).

4.2.1.3. *Inhibin and activin*

Two other hormones are also involved in the control of male reproduction: inhibin and activin. Both are produced by the Sertoli cells in response to total sperm production, and have additional feedback effects on hypothalamic and pituitary function, specifically on FSH production. Inhibin acts as a negative and activin as a positive feedback (Roser, 1997). The precise role of inhibin and activin in the horse is as yet largely unclear.

4.2.1.4. *Prolactin*

The central role that prolactin may play in translating day length into non-reproductive seasonal physiological changes has been discussed in this chapter and in Chapter 3. In addition, based upon work in other farm livestock, prolactin may also have a role in enhancing the effect of LH on the Leydig cells in some way and on the functioning of the accessory glands (Thomson *et al.*, 1996). The importance of prolactin specifically in the horse is as yet unclear.

Fig. 4.7. Concentration (mean + standard error) of testosterone in the peripheral plasma of mature stallions over a 13-month period (from Berndston *et al.*, 1974).

4.2.1.5. Oestrogens

The stallion's testis contains a higher concentration of oestrogens and oestrones (150–200 pg ml⁻¹) than the testis of other mammals. The control and significance of such testicular oestrogen are unclear (Raeside, 1969; Seamens *et al.*, 1991; Amann, 1993b; Landeck, 1997).

4.3. Behavioural Changes

Testosterone drives male behaviour, especially that associated with reproduction. There is much variation between individuals, but in summary the following generalized behaviour is controlled by testosterone (Pickett *et al.*, 1975a). On sight of a mare, the frequency and amplitude of GnRH and hence LH and FSH release increase, driving testosterone production and hence producing stallion behaviour (Irvine and Alexander, 1991). In addition, 20% of GnRH released acts directly on higher brain centres (Merchenthaler *et al.*, 1989; Pozor *et al.*, 1991). Behavioural patterns of a stallion include fixation of his eyes upon the mare, neck arching, stamping or pawing the ground and general elevated stallion stance. He will often show the characteristic flehman behaviour of drawing back his top lip as if tasting the air, accompanied by roaring. If the mare seems receptive, he will approach her from

the front, muzzle to muzzle, and in the absence of hostility will work his way over her neck, back and rump towards her perineum and vulva. If the mare still stands with no objection, he will turn and approach her from behind and to one side, nearly always the left-hand side. He will then mount her from that side, possibly after a few initial dummy mounts to test her reaction and confirm that she is willing to stand.

Ejaculation follows shortly after entry into the mare and is signalled by the rhythmic flagging of the tail. Such mating behaviour is directly affected by circulating testosterone concentrations. At the beginning and the end of the season, reaction time to a mare is longer and the number of mounts per ejaculate is greater, these being direct indications that, when testosterone levels are declining, sexual enthusiasm or libido is also waning (Figs 4.4 and 4.5; Weber and Woods, 1993).

4.4. Conclusion

The control of reproduction in the stallion is designed to ensure continual reproductive activity, rather than cyclic as in the mare. The only constraint on the stallion's reproductive activity is season, a limitation that ensures that offspring are more likely to be born at a time of year most appropriate to their survival.

5

The Anatomy and Physiology of Pregnancy in the Mare

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5.1. Introduction

The anatomy of pregnancy in the mare can be divided into four main sections for ease of consideration: fertilization; early embryo development; placentation; and organ growth. Further, more detailed accounts may be found in other texts, such as Douglas and Ginther (1975), Betteridge *et al.* (1982), Flood *et al.* (1982), Ginther (1992), Asbury and Le Blanc (1993) and Flood (1993).

5.2. Fertilization

The ovum released by the follicle is directed down the Fallopian tube by the fimbriae lining it to where it waits in the ampulla region, that

nearest the infundibulum, for the arrival of the sperm. It is unable to pass through the utero-tubular junction until it has been fertilized.

The sperm, having been ejaculated into the top of the cervix/bottom of the uterus, make their way up through the uterus to the utero-tubular junction. They move by means of contractions of the female tract and the driving action of their own tails. It is thought that they are attracted towards the ovum by chemical attractants produced by the ovum awaiting fertilization. On arrival at the utero-tubular junction, they pass through to the Fallopian tube and, if the timing is correct, meet a waiting ovum in the ampulla. As the sperm pass up through the female tract, they come in contact with uterine secretions, which induce a capacitation response in the acro-

some region of the sperm heads, a response that is essential before it is capable of fertilizing an ovum. Capacitation activates the enzymes within the acrosome region in readiness for penetration. Once in the vicinity of the ovum, sperm stick to the outer gelatinous layer, this layer having replaced the corona radiata cells prior to ovulation (Fig. 5.1).

Sperm, by means of the whipping action of their tails, force themselves through the outer gelatinous layer to the zona pellucida. They then penetrate the zona pellucida, using the enzyme acrosin released from the sperm head of capacitated sperm. This enzyme digests a pathway through the zona pellucida.

As the sperm head meets the vitelline membrane of the ovum, the two fuse. This fusion initiates the final meiotic division, resulting in three polar bodies and the single ovum nucleus. The nuclei of the sperm and the egg (often termed the pronuclei) unite, their haploid complement (32) of chromosomes joining together to give the full diploid (64) of the new individual. This newly combined genetic material now dictates all the characteristics of the new individual.

There is some variation in the reported length of time that the equine ovum remains viable; figures varying between 4 and 36 h have been reported, though there is one report of fertilization occurring 7 days after ovulation (Newcombe, 1994). After fertilization, the ovum is known to actively control, possibly via the localized secretion of oestro-

gens or prostaglandin E, its passage through the utero-tubular junction to the uterine horn, overtaking on its way any unfertilized ova from that or previous ovulations. Any ova not fertilized may take several months to degenerate (Betteridge and Mitchell, 1975; Onuma and Ohnami, 1975; Flood *et al.*, 1979a,b; Ball and Brinsko, 1992).

In order to ensure the successful fusion of one male pronucleus and one female pronucleus, it is essential to ensure that only one sperm penetrates the vitelline membrane of the ovum. Polyspermy (penetration by more than one sperm) is prevented by an instantaneous block, which occurs as soon as one sperm touches the vitelline membrane. This instantaneous response involves a chemical reaction within the vitelline membrane, forcing a gap between itself and the zona pellucida. No sperm can cross this gap and hence an instantaneous block to polyspermy is ensured.

5.3. Early Embryo Development

Twenty-four hours after mating, the fertilized ovum, now termed a zygote, has divided by mitosis (growth by cell division) into two cells. At this stage, the outer gelatinous layer is lost and the fertilized ovum, still within the zona pellucida, continues to divide into 4, 16, 32 cells, etc. At 4 days old, it is a bundle of cells, again still contained within its zona pellucida. It is now termed a morula (Fig. 5.2).

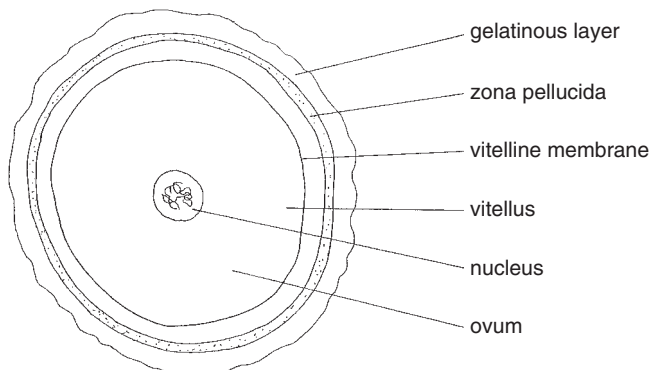


Fig. 5.1. The equine ovum prior to ovulation.

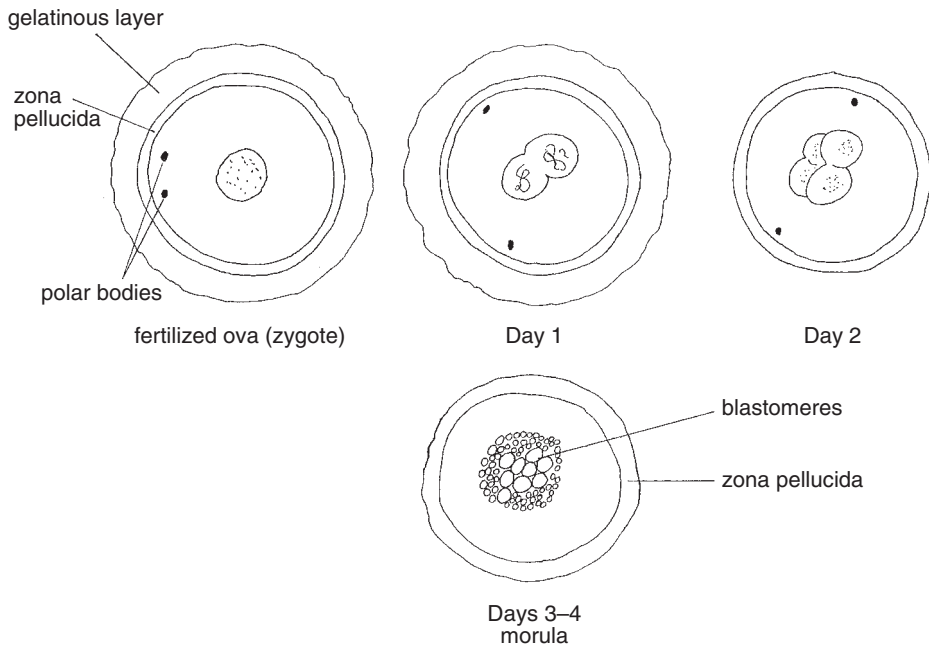


Fig. 5.2. The developmental stages from fertilized ovum to morula in the equine conceptus, illustrating the loss of the gelatinous outer layer by day 2.

At this stage the total volume and external size of the bundle of cells have not changed from the two-cell zygote stage. The cytoplasm of the original ovum has either been divided up between all the cells in the morula or used for energy. Nevertheless, the amount of genetic material has dramatically increased, giving a full identical complement to all cells of the morula.

As the cells continue to divide, the morula makes its way towards the uterotubular junction by anticlockwise rotational swimming. It passes through this junction and arrives in the uterus at day 5–6. At day 5, a thin acellular glycoprotein layer, termed the capsule, appears in the perivitelline space between the trophoctoderm and the zona pellucida (Oriol, 1994). The function of this capsule is unclear. It may have a role in preventing the adhesion of the embryo to the endometrium, hence allowing the prolonged mobility phase characteristic of equine conceptuses. It may also have a role in driving embryo expansion, which occurs from day 5 (Crossett *et al.*, 1995;

Fig. 5.3). From day 6, the total size of the embryo starts to increase; this helps to force the thinning of the zona pellucida, which eventually breaks. The embryo then hatches through this break and is left surrounded by its capsule. At this time, the conceptus starts to derive nutrients for its growth and cell division from the surrounding uterine secretions, as by this stage it has used up all its own reserves. These nutrients are passed through the capsule to the embryonic cells via carrier proteins (Stewart *et al.*, 1995). The provision of such additional nutrients allows a further increase in size. The morula is now in its mobility phase, free and floating within the uterus, deriving all its nutritional requirements from uterine histotroph (milk), a secretion designed to match exactly the requirements of the developing conceptus.

At day 8, the cells of the morula become differentiated (organized) and three distinct areas can be identified: the embryonic disc (shield or mass); the blastocoel; and the trophoblast (Fig. 5.4). The morula is now termed a blastocyst.

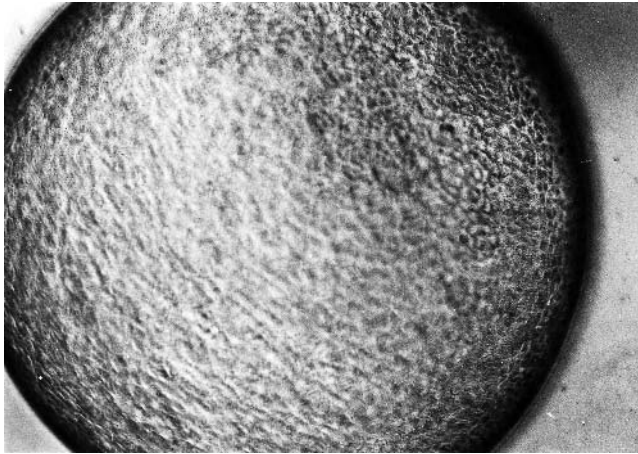


Fig. 5.3. A view of the surface of an equine embryo illustrating the outer trophoblastic cells. The capsule is evident as a clear area encircling the whole conceptus (photograph Alison Crook).

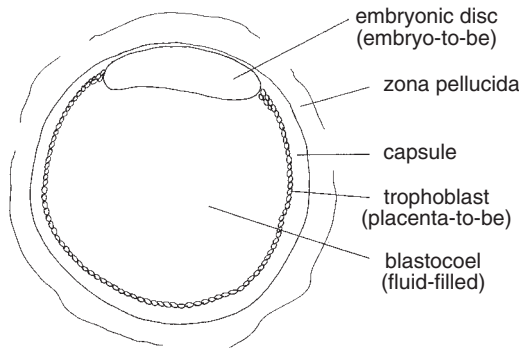


Fig. 5.4. The equine blastocyst at day 8 postfertilization, showing the differentiation of three areas.

These three areas go to form the embryo proper (embryonic disc), the yolk sac (blastocoel) and the placenta (trophoblast). This cell differentiation marks the beginning of the switching on and off of various genes, cells then becoming destined to pursue set lines of development. Prior to this differentiation, all cells in theory are capable, if extracted from the morula, of each developing into a new individual, as none of its genes has been switched off. After differentiation, this is no longer possible, as certain cells have been given the message to pursue only set lines of development. The mechanism behind this switching on and off of genes and its trigger are unknown in the horse. It is important to

note that, at this differentiation stage, the conceptus is very susceptible to external physical effects, e.g. drugs, other chemicals, disease, radiation, etc. These can disrupt the differentiation process, resulting in deformities, abnormalities and a high risk of abortion or reabsorption.

From this stage further differentiation takes place. Day 9 marks the differentiation of two germ layers (cell layers): the ectoderm, consisting of the outer blastocyst cell layers; and the endoderm, consisting of the inner cell lining (Fig. 5.5).

The endoderm grows and develops, working its way around the inside of the trophoblast to give a complete inner layer. The

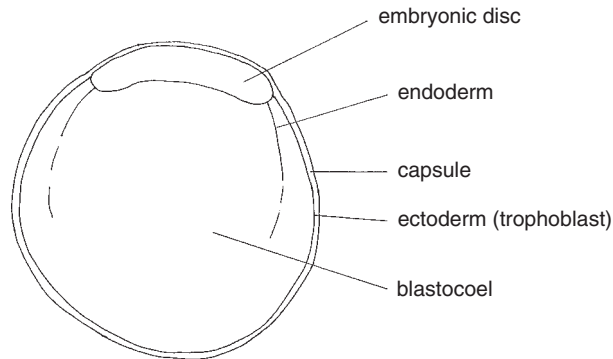


Fig. 5.5. The equine conceptus at day 9 postfertilization, illustrating the differentiation of the ectoderm and endoderm layers.

endoderm and ectoderm together form the yolk sac wall and provide the means by which the embryonic disc receives its nourishment from the uterine secretions. The blastocoel, or fluid-filled centre, is now termed the yolk sac and acts as a temporary nutrient store (Fig. 5.6). This remains the major source of nutrients to the embryo until implantation or fixation occurs. The equine embryo is unique in being free living within the uterus for up to 25 days prior to final implantation; this period of time is termed the mobility phase.

At day 14, when the embryo has reached 1.3 cm in diameter, the mesoderm or third germ-cell layer begins to develop. It becomes progressively evident between the ectoderm and endoderm, in the centre of the yolk sac wall, again working its way down from the embryonic disc to enclose the whole blasto-

cyst. These three germ-cell layers are the cell layers from which all placental and embryonic tissue development originates. In the case of the placenta, the ectoderm forms the outer cell layers nearest the uterine epithelium, the mesoderm forms the blood vessels and nutrient transport system and the endoderm forms the inner cell lining, which will become the allantoic sac (Fig. 5.7).

At day 16, folds appear in the outer cell layers and the beginnings of the protective layers, which will surround the embryo, become evident. The ectoderm folds over the top of the embryonic disc, taking the mesoderm with it. The outer layer of these folds is now made up of the ectoderm plus a mesoderm layer and is termed the chorion. These two folds fuse, producing a fluid-filled protective space for the embryonic disc; this is

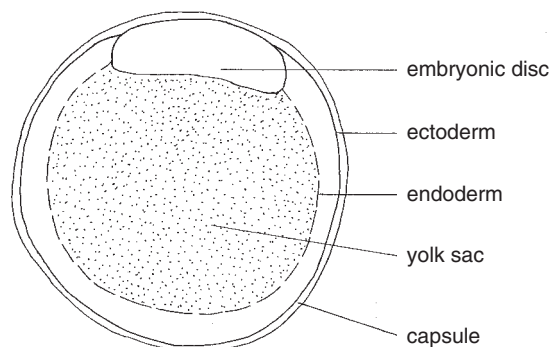


Fig. 5.6. The equine conceptus at day 12 postfertilization, illustrating the yolk sac, which at this stage provides the nutrients required by the developing conceptus.

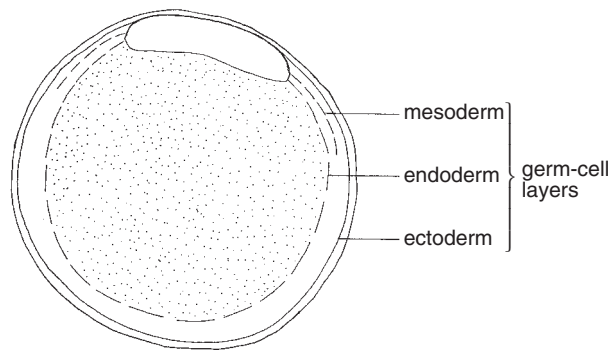


Fig. 5.7. The equine conceptus at day 14 postfertilization, illustrating the developing mesoderm, which forms the blood vessels and nutrient transport system of the conceptus.

the amniotic sac containing the amniotic fluid (Fig. 5.8). At this stage, the first fixation of the embryo to the epithelium is reported to occur, though this attachment may be only temporary (Waelchi *et al.*, 1996).

The membrane encompassing the amnion and separating it from the surrounding allantoic fluid (discussed later) is termed the allantoamniotic membrane. Initially, the amnion is visible as a clear fluid-filled bubble surrounding the embryo. As pregnancy progresses, it tends to collapse and lie close to the fetus. Throughout its life *in utero*, the amniotic sac provides a clean and protective environment in which the embryo can develop. The source of its surrounding amniotic fluid is not clear. However, its composition is very much like blood serum, and exchange of fluids between the amniotic sac and the kidneys, intestine and respiratory

tract is known to occur. The fetus in later stages seems to breathe in and swallows its surrounding amniotic fluid. The volume of amniotic fluid surrounding the fetus is about 0.4 l at 100 days postfertilization and increases to 3.5 l at full term.

At day 16, when the amnion is first evident, the mesoderm has not yet spread to enclose the whole of the yolk sac. The area over which the mesoderm has spread, and which, therefore, has the three layers, ectoderm, mesoderm and endoderm, is nearest to the embryonic disc, is called the trilaminar omphalopleure. The area into which the mesoderm has not yet spread and which has only ectoderm and endoderm is termed the bilaminar omphalopleure. The junction of these two areas, i.e. the line delineating the limit of mesoderm migration, is called the sinus terminalis (Fig. 5.9).

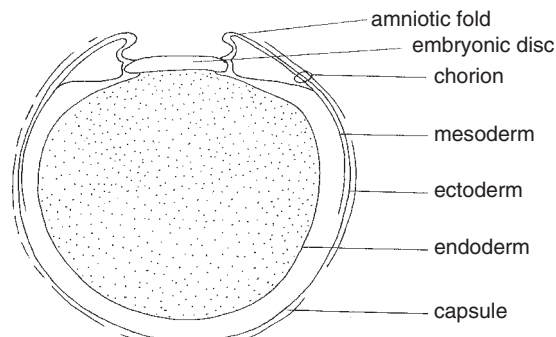


Fig. 5.8. The equine conceptus at day 16 postfertilization, illustrating the formation of the amniotic folds over the embryonic disc.

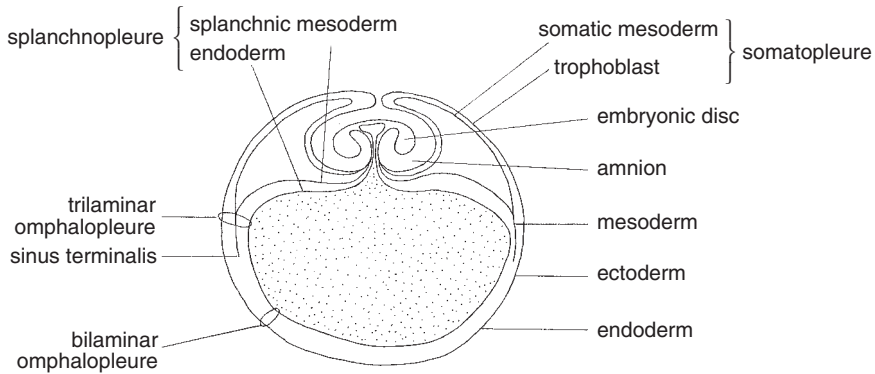


Fig. 5.9. The equine conceptus at day 18 postfertilization, illustrating the near-completion of the amniotic sac surrounding the embryonic disc. The trilaminar omphalopleure is shown, nearest the embryo, consisting of the endoderm, mesoderm and ectoderm, and the bilaminar omphalopleure, into which the mesoderm has not yet spread.

From this stage, it is increasingly evident that embryology can be dealt with in two sections: placentation; and organ development.

5.4. Placentation

The placenta has two major functions: first, protection; and, second, regulation of fetal environment, in the form of nutrient intake and waste output. The placenta develops from the extraembryonic membranes, the trophoblast of the blastocyst. The first source of nutrients, and therefore a form of primitive placenta, is the yolk sac or blastocoel. This provides both a temporary store and a transport system for nutrients derived from uterine secretions.

Day 14 sees the first evidence of blood vessels developing in the centre of the yolk sac wall in the mesoderm. This will become the blood system of the placenta. By day 18, the vitelline artery, carrying blood towards the mother, and the vitelline vein, carrying blood away from the mother, are identifiable.

On day 20, an outpushing of the embryonic hind-gut can be seen immediately below the placenta. This is termed the allantois and it continues to grow with the embryo. This sac is filled with allantoic fluid; it is encompassed by the allantochorionic membrane, or placenta. The allantoic fluid consists

of the secretions of the allantochorion, along with urinary fluid, which is excreted from the fetal bladder via the urachus within the umbilical cord (Figs 5.10 and 5.11).

The volume of the allantois at day 45 is approximately 110 ml, increasing to 8.5 l by day 310, a considerably larger volume than seen in the amniotic sac. The allantoic fluid increases in volume as the fetus grows, producing more urinary fluid to be stored. During the first trimester (3–4 months), it is clear yellow in colour, changing to brown/yellow as pregnancy develops (Figs 5.12 and 5.13).

This developing allantoic sac moves over the top of the embryo as its contents increase, forcing the embryo down to the bottom of the blastocyst, reducing, as it goes, the extent of the yolk sac, until the yolk sac is hardly visible. As the allantoic sac increases in size, the umbilical cord becomes evident. The attachment point of the umbilical cord normally corresponds to the position of initial implantation. It consists of two vitelline arteries, one vitelline vein and the urachus, plus some supporting and connective tissue. The arteries and veins are responsible for blood transfer to and from the placenta to the fetal system and the urachus transfers waste products from the bladder to the allantois; as such, it extends no further than the allantois and does not reach the placenta (Fig. 5.14).

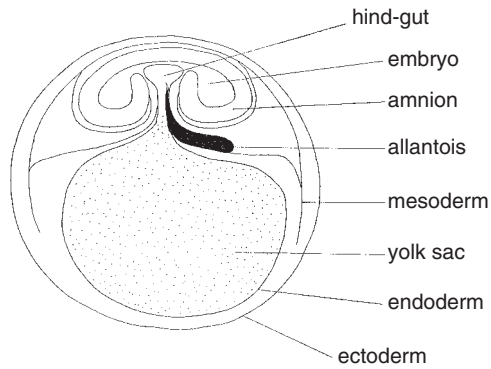


Fig. 5.10. The development of the equine placenta at day 20 postfertilization, illustrating the development of the allantoic sac.

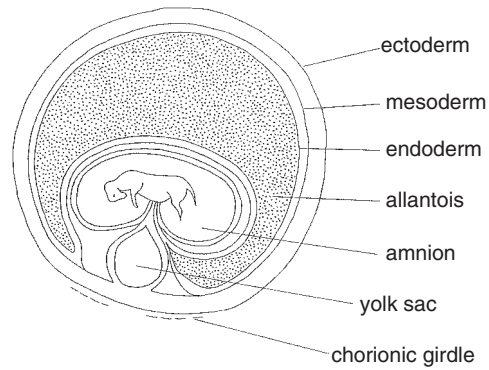


Fig. 5.11. The development of the equine placenta at day 40 postfertilization.



Fig. 5.12. The equine fetus at 200–220 days of gestation, illustrating the removal of the allanto-chorion (placenta) and the significant amount of allantoic fluid released as a result.



Fig. 5.13. Equine fetus at 200 days of gestation, illustrating the fetus within the amnio-chorion after removal of the allanto-chorion. The umbilical chord can be seen connecting the fetus and the allanto-chorion, which has been removed from around the fetus and is lying in the top left-hand corner. The, as yet small, hippomane can be seen just above the ruler.

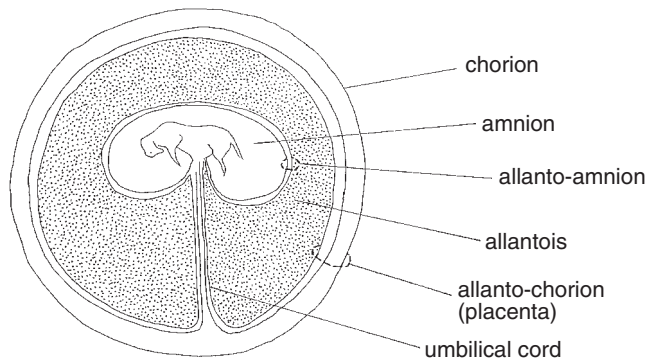


Fig. 5.14. The placental arrangement of the equine conceptus near term.

As the fetus develops, its nutrient demand increases. The nutrients provided via the yolk sac are soon not enough to meet this demand; thus a more intimate relationship needs to develop between the mother and the embryo, and hence its period of mobility ceases and it begins to implant. This occurs as a gradual process from day 25 onwards; until this point, the yolk sac is fully functional. At this stage, the capsule begins to degenerate, though remnants have been reported as late as day 35 (Enders *et al.*, 1993; Oriol, 1994).

The first identifiable attachment between mother and fetus occurs around day 25, at the chorionic girdle (Fig. 5.15). This is a temporary attachment and normally implants the conceptus at the junction between the uterine body and the uterine horn. The chorionic girdle is a band of shallow folds encircling the allanto-chorionic membrane. Cells within this girdle elongate and invade the uterine endometrium, engulfing some of the epithelial cells. This girdle forms in an area of the conceptus where there is no mesoderm, i.e. where the yolk sac is being restricted by the

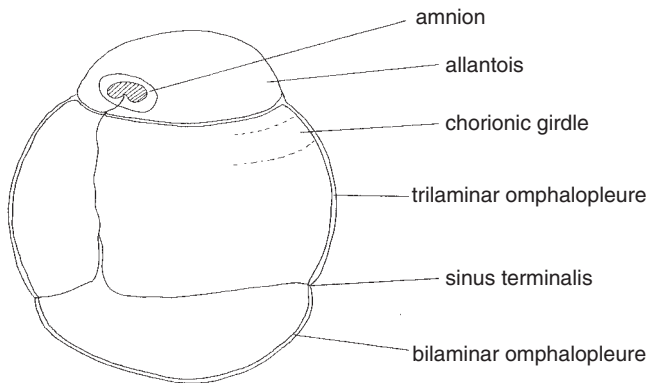


Fig. 5.15. The beginnings of the development of the placenta of the equine conceptus at day 25 postfertilization, illustrating the position of the chorionic girdle attachment.

developing allantois. The growth of the chorionic girdle is likely to be due in part to insulin growth factor II (Enders and Lui, 1991; Enders *et al.*, 1993). This attachment, though only tenuous, does provide a significant exchange unit (Enders *et al.*, 1993). At day 38, the fetal cells migrate into the maternal endometrium and detach from the allanto-chorion. Some work suggests that the conceptus is then released and can migrate within the uterus. However, it normally returns to the junction of the uterine horn and body for final implantation around day 40, though the exact time may vary, final fixation

possibly occurring later in older mares (Carnevale and Ginther, 1992). The invading fetal girdle cells now form endometrial cups in a band around the inside of the uterus, at the junction of the body and the horn, at the original position of the chorionic girdle (Fig. 5.16). Their development is associated with increasing lymphatic activity (Enders and Lui, 1991). These endometrial cups secrete equine chorionic gonadotrophin (eCG), sometimes referred to as pregnant mare serum gonadotrophin, which is essential for the maintenance of early pregnancy. eCG will be discussed in detail in Chapter 6.

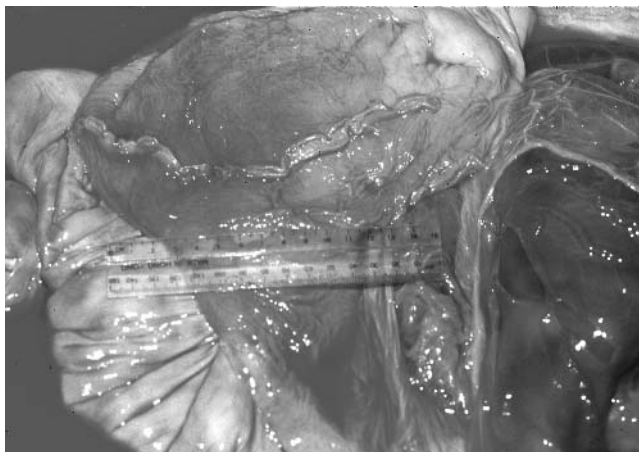


Fig. 5.16. The remains of the endometrial cups can be seen in a band running across the uterine endometrium.

Around day 90, the endometrial cups begin to degenerate and slough away from the uterine endometrium. The reason for this seeming rejection is not fully understood, but it may be a maternal immunological rejection of the 'foreign' fetal tissue, which is specific to the endometrial cups and not the allanto-chorion (Asbury and Le Blanc, 1993). The duration of the endometrial cups is very variable, being longer in sibling matings, primiparous mares (mares not previously pregnant) and foal-heat matings (Spincemaille *et al.*, 1975; Bell and Bristol, 1991; Allen *et al.*, 1993). The remains of these sloughed-off endometrial cups may be re-absorbed by the fetus during the remainder of the pregnancy or they may be seen in the placenta at birth as invaginations or pouches in the allanto-chorion (Fig. 5.16).

Gradually, over time, as the endometrial cup attachment is lost, the rest of the fetal allanto-chorion begins to attach to the uterine epithelium. This attachment begins between day 45 and day 70 and gradually becomes firmer over the next 100 days, being fully attached by day 150. At day 45–70, the allanto-chorion takes on a velvety appearance, created by fine microvilli over its entire surface; hence the equine placenta is termed diffuse. These microvilli organize themselves into discrete microscopic bundles or tufts, which invade receiving invaginations in the uterine epithelium. These bundles of

microvilli are termed microcotyledons, and their attachment develops over a period of time, being fully complete and functional by day 150 (Fig. 5.17).

A strong attachment is formed between the fetus and the mother. The equine placenta is relatively thick, with six cell layers and four basement membranes. The three cell layers on the fetal side are: mesoderm (endothelium – blood vessel wall); endoderm (connective tissue); and ectoderm (allanto-chorion); and the three on the maternal side are: epithelium; endometrium (connective tissue); and endothelium (blood vessel wall). The equine placenta is therefore termed epitheliochorial and covers the whole surface of the uterus, except the cervix (the cervical star) and the two utero-tubular junctions (Figs 5.18 and 5.19; Figs 5.12 and 5.13; McDonald and Fowden, 1997).

The presence of the microcotyledons serves to increase the surface area of the placenta and, therefore, the area for nutrient and gas exchange. Within each microcotyledon, the maternal and fetal blood-supply system come in close proximity, allowing efficient diffusion.

However, the thickness of the placental attachment prevents the diffusion of any large protein molecules. As immunoglobulins are large protein molecules, the attainment of passive immunity in the foal by

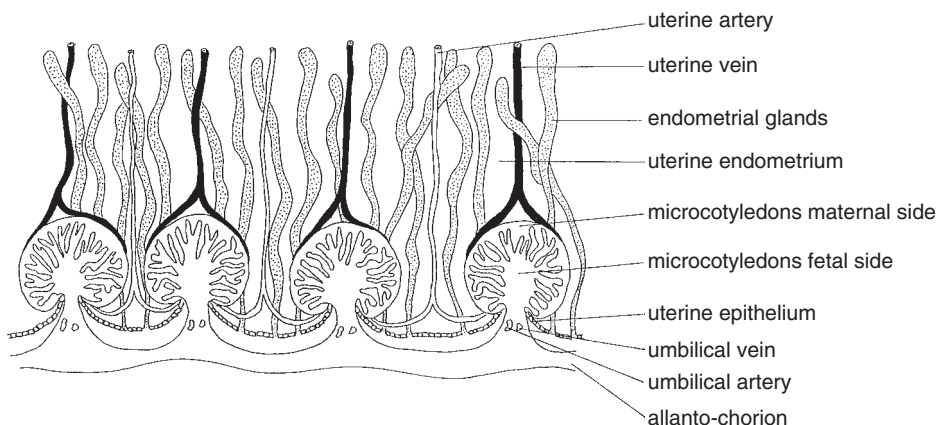


Fig. 5.17. Equine placental microcotyledons in the fully developed placenta.

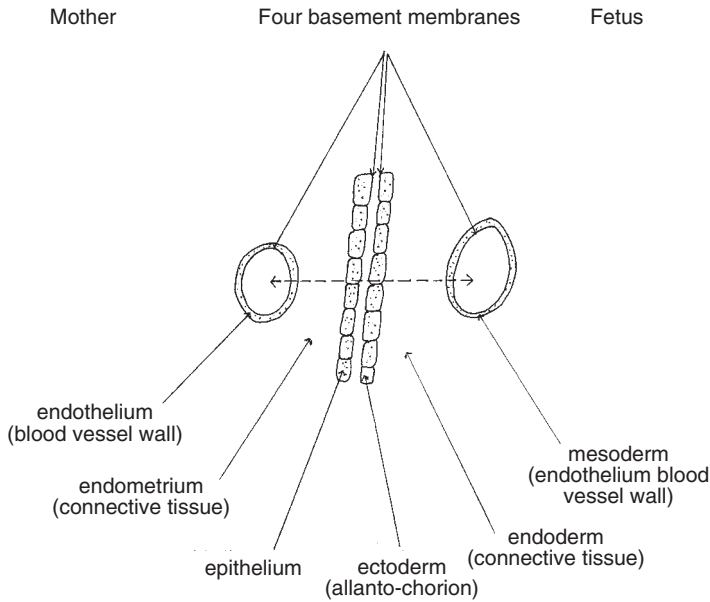


Fig. 5.18. The structure of the equine placenta.

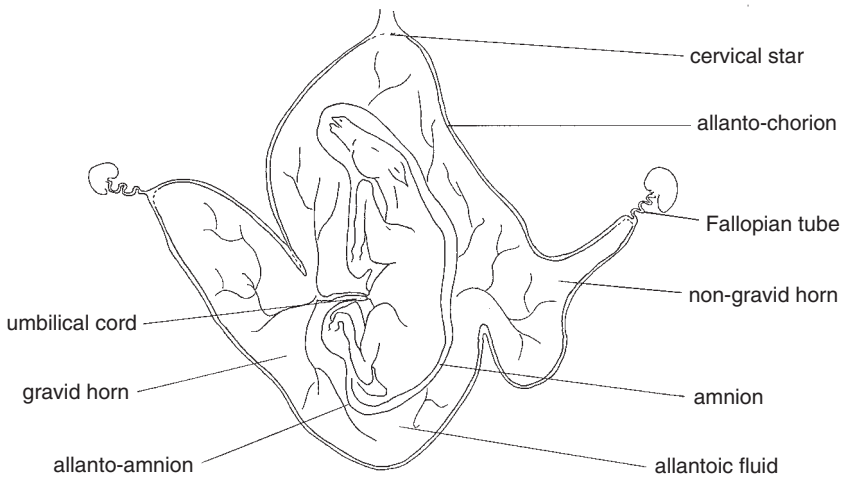


Fig. 5.19. The equine fetus and placenta near term.

diffusion across the placenta is limited. Passage of immunoglobulins via colostrum is, therefore, of utmost importance in the mare, as will be discussed in further detail in Chapters 15 and 16. The thickness of the placenta varies in other mammals, but, in general, the thicker the placenta, the less

efficient the transfer of passive immunity *in utero* and hence the increased reliance upon colostrum. However, a thicker placenta, as seen in the mare, has the advantage of providing extra protection to the fetus from harmful maternal blood-borne factors (Silver *et al.*, 1973).

5.4.1. Placental efficiency

Despite the thickness of the placenta, nutrient and gaseous exchange across the mare's placenta is relatively efficient when compared with other farm livestock. This is accounted for by the diffuse nature of the equine placenta compared with the cotyledonary (attachment in discrete areas) nature of those of the ewe and the cow. However, it must be remembered that measurements taken on placental efficiency involve the acute catheterization of the umbilical arteries, and hence the technique used may affect the results obtained. Silver *et al.* (1973) demonstrated the relative efficiency of the mare's placenta, as changes in maternal blood oxygen and glucose concentrations were mimicked more closely by changes in the fetal blood concentration than in sheep. Free fatty acids and lactate also follow the same pattern. It may well be deduced, therefore, that factors affecting the mare will have a greater effect on the fetus than is evident in ruminants, though such an association has yet to be confirmed.

As pregnancy progresses, the maternal epithelium stretches as the uterus increases in size. As a result, the placenta also stretches and becomes thinner and hence the resistance to gaseous and nutrient exchange decreases, the placenta becoming more efficient as the demands of the fetus increase. By full term, the placenta of a 15–16 hh horse weighs about 4 kg. Its surface area is approximately 14,000 cm² and it is about 1 mm thick. The foal's birth weight is directly proportional to the surface area of the placenta, as this is the limiting factor controlling nutrient and gas exchange and hence their availability to the developing fetus. The surface area of a placenta may be restricted for several reasons, including the presence of twins.

Thoroughbred. The incidence of twin ovulations, which have the potential to result in twin conceptuses, in the Thoroughbred is 20–25% (Davies Morel and O'Sullivan, 2001). Of this potential number of twins, significant natural reduction does occur, with one twin dying (70% of twins are unilateral, of which 85% naturally reduce, and 30% are bilateral, none of which naturally reduces) (Ginther, 1989a,b; Ginther and Griffin, 1994). If twins do develop to the placentation stage, the area of the uterus available for each placenta is restricted by the presence of the other fetus. If the division of uterine surface area available to each twin is equal, then both twins have an equal chance of survival, but their birth weights will be reduced. If the division is unequal, then the smaller one may cause the whole pregnancy to abort or, if the pregnancy is not well advanced, it may die and become mummified. If mummification occurs, the pregnancy may well continue, but the placenta of the larger surviving fetus cannot expand into the uterine surface originally occupied by the now dead fetus (Fig. 5.20). At term, therefore, a single foal will be born, but with a reduced birth weight due to placental restriction (Fig. 5.21).

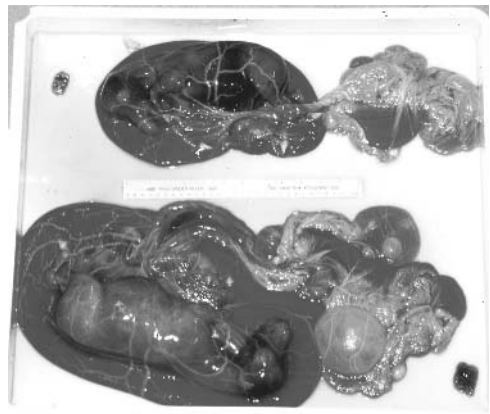


Fig. 5.20. The fetuses of a twin pregnancy dissected out at *post mortem*. The different size of the twins is evident as a result of placental restriction of the smaller twin. If left to go to term, the smaller twin would eventually have died and probably have caused the abortion of the whole pregnancy.

5.4.2. Twins

Twinning is increasingly a significant problem in stud management, especially in intensively bred horses, such as the

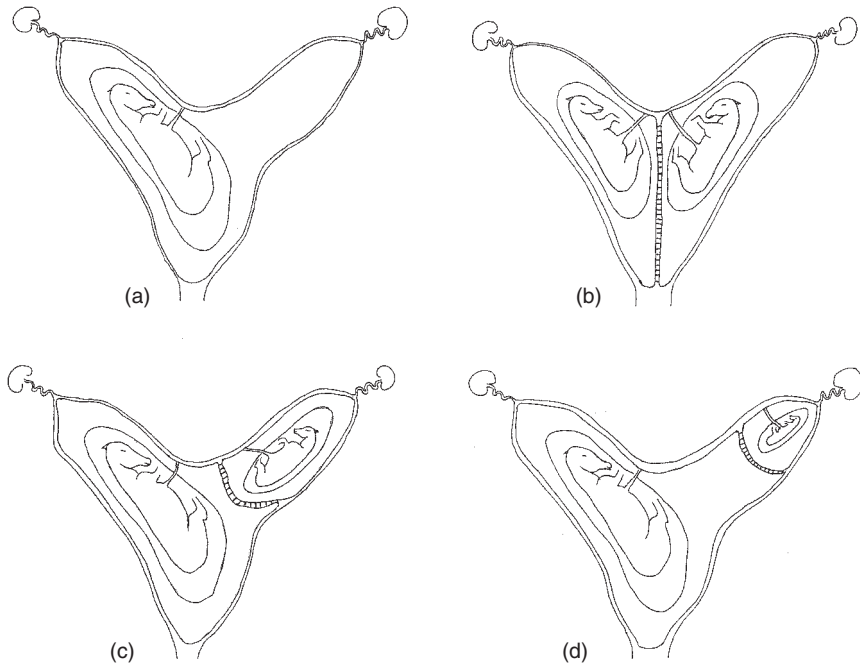


Fig. 5.21. Placental configurations in equine singleton and twin pregnancies: (a) singleton; (b) equal split, bilateral (50% : 50%); (c) unequal split, bilateral (60% : 40%); (d) unequal split, bilateral (80% : 20%).

5.4.3. Placental blood supply

As mentioned previously, the mesoderm of the blastocyst surrounding the yolk sac forms the first blood supply to the fetus. Early on, a clear network of blood vessels can be identified within the trilaminar omphalopleure, with two major vessels connecting the network to the rudimentary heart. Additional pathways develop to feed areas of considerable growth. Hence, when the yolk sac degenerates and the nutrient supply to the fetus is taken over by the allanto-chorionic placenta, a well-formed network of blood vessels lining the allanto-chorion already exists. This fine network enlarges and invaginates into the microcotyledons of the placenta. Each microcotyledon is supplied on the fetal side by several arteries, but exit back to the fetal heart is via a single vein. This arrangement slows down the flow of blood through the microcotyledons and encourages more efficient diffusion

and gas exchange. The oxygenated and nutritionally replenished blood returns to the fetal heart by the umbilical vein (Fig. 5.22). The umbilical cord, therefore, contains two fetal arteries and one fetal vein plus the urachus.

It should be remembered that in the fetus, because of the bypass of the non-functional lungs, deoxygenated blood is carried to the placenta in arteries and oxygenated in the veins.

On the maternal side a very similar arrangement exists. Oxygenated and nutritionally enriched blood approaches the microcotyledons in a fine network of arteries, but the drainage back to the maternal system is also via a single vein, again slowing down the passage of blood and increasing the efficiency of nutrient and gaseous exchange. This transfer across the utero-fetal placental barrier can be compared in many ways to the gaseous exchange within the mammalian lung.

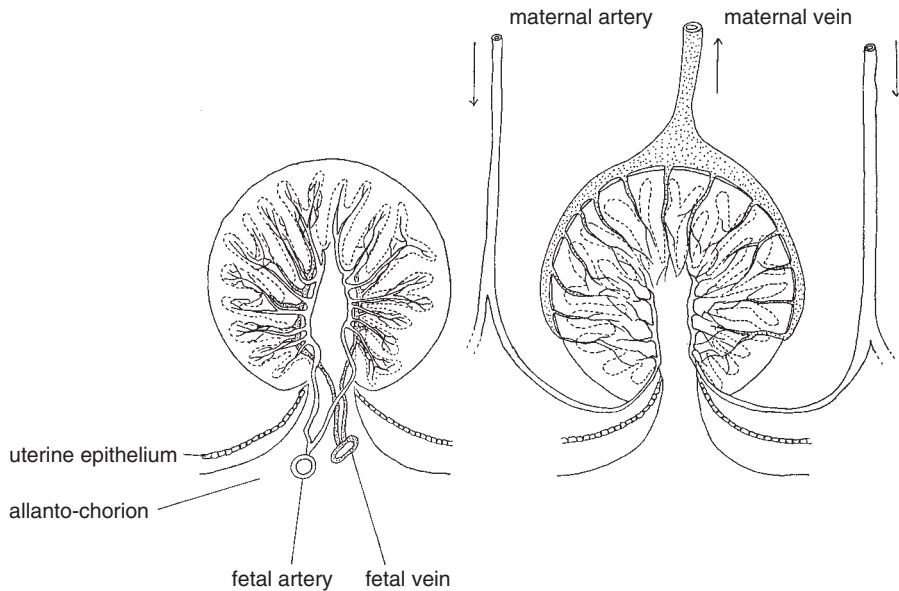


Fig. 5.22. The venous and arterial blood supply to the microcotyledons within the equine placenta, allowing the transfer of nutrients and waste products from the maternal (right) to the fetal (left) system and vice versa.

5.5. Organ Development

Organ development arises from the reorganization of cell populations within the embryonic disc itself. This organization is related to that which occurs in placentation, previously discussed. This can be divided into two basic sections, gastrulation and neurulation. The former can be subdivided into segregation, delamination and involution. Further accounts of organ development can be found in Douglas and Ginther (1975), Van Niekerk and Allen (1975), Betteridge *et al.* (1982), Flood *et al.* (1982) and Enders *et al.* (1988).

5.5.1. Gastrulation

Gastrulation is defined as the organization of the embryo into three germ layers: ectoderm; mesoderm; and endoderm. This involves primarily the cells of the embryonic disc but also those of the placental tissue. The first stage of gastrulation is segregation, during which the central blastomeres or cells of the embryonic disc organize themselves into smaller outer and larger inner blastomeres (Fig. 5.23).

The larger blastomeres collect underneath the disc and migrate in two directions. First, they migrate to line the remaining ectoderm of the blastocyst, forming the endoderm. Second, they migrate within the embryonic disc, creating at day 11 the first asymmetry, a thicker area at the caudal end and a thinner area at the cranial end (Fig. 5.24).

The next stage of gastrulation is termed delamination. This commences at day 12 and marks the first evidence of epiblast cells, hypoblast cells and the primitive gut (Fig. 5.25).

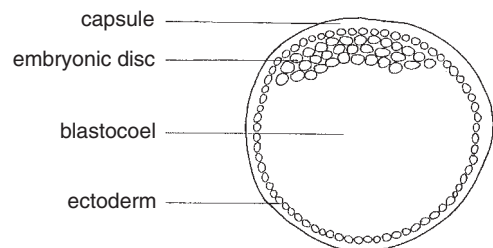


Fig. 5.23. Day 9 segregation in the equine conceptus, illustrating the larger inner blastomeres and smaller outer blastomeres within the embryonic disc.

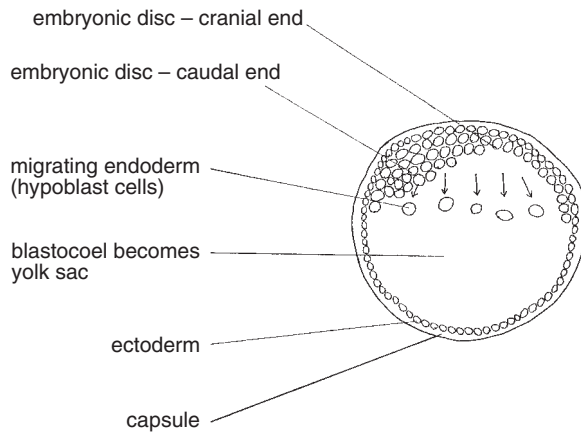


Fig. 5.24. Day 11 segregation in the equine conceptus, indicating the migration of the large blastomeres from the lower part of the embryonic disc to form the ectoderm along with the first asymmetry in the embryonic disc.

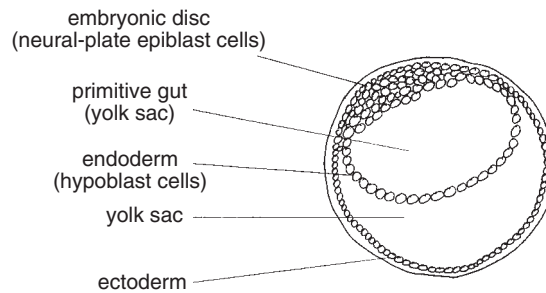


Fig. 5.25. Day 12 delamination in the equine conceptus, showing the future development of the endoderm layer.

The epiblast cells are those of the embryonic disc. The hypoblast cells are the migrating endoderm and within this ring of hypoblast cells is the yolk sac or primitive gut. At day 14, a change in this neural plate becomes evident. This change forms the beginning of the primitive streak identified within the epiblast cells. At this stage, it is about 1 cm in length.

Involution is the third stage of gastrulation, when the epiblast cells move inwards to the centre of the caudal end of the disc (Fig. 5.26). At this stage, three types of cells – ectoderm (epiblast cells), mesoderm and endoderm (hypoblast cells) – are evident within the embryonic disc as seen in the extra embryonic tissue (Fig. 5.7). These three cell layers will go to form all the main body structures.

The moving ectoderm or epiblast cells reappear as mesoderm between the ectoderm and the hypoblast cells or endoderm. As the cells move through to the lower level, they leave a depression in the upper surface of the epiblast. These migrating epiblast cells tend to move in greater concentrations at the caudal end of the primitive streak, making it wider. The primitive streak so formed makes the future longitudinal axis of the embryo.

At day 15, epiblast cell movement tends to slow down; the slight indentation along the longitudinal axis of the primitive streak becomes deeper, as cells continue to move out from underneath to form the mesoderm and are not replaced by migrating epiblast cells above. This deep groove is now termed the primitive groove. The cells associated

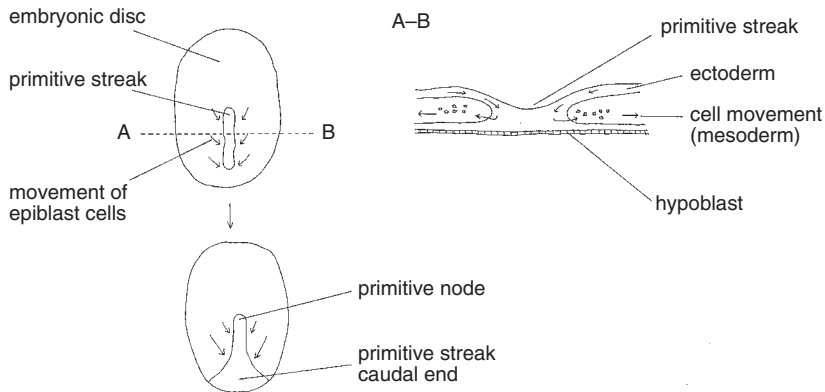


Fig. 5.26. Day 14 involution of the equine conceptus. A bird's-eye view of the embryonic disc, along with a cross-section view through A–B, illustrating the passage of ectoderm cells through the primitive streak to reappear between the ectoderm and endoderm, forming the mesoderm. Further cell movement results in the flattening of the caudal end of the primitive streak.

with the primitive groove are termed node cells, to differentiate them from the cells of the remainder of the embryo. At day 15, these node cells can be identified as precursors of future body organs. The ectoderm node cells form the neural plate, running the length of the top of the primitive groove, the cranial end of which goes to form the head. The spreading mesoderm in the immediate vicinity of the neural plate goes to form the somites, or body trunk, and the mesoderm immediately below the primitive groove goes to form the notochord (spine and central nervous system (CNS)). Finally, the wide caudal end forms the tail end of the fetus (Fig. 5.27).

The process of gastrulation is now completed, the major cell blocks are identifiable and the longitudinal axis of the embryo is determined.

5.5.2. Neurulation

The next stage, termed neurulation, involves the development of the CNS, gut and heart. Day 16 sees three major changes. First, the ectoderm near the neural plate thickens and two neural folds develop either side of the neural plate. The neural plate becomes depressed and the neural folds fold over, join and then fuse to enclose a hollow tube, the spine- and CNS-to-be (Fig. 5.28).

Second, the mesoderm either side of the neural plate organizes itself into 14 somites. Third, at the cranial end of the neural plate, an increase in cell growth above the surface becomes apparent, with an accompanying increase in the length of the neural plate. This cell growth folds over to form the head process, heart and pharynx.

By day 18, lateral folds are beginning to develop either side of the head process. As cells move into this area and cell division increases, the cranial end of the neural plate lifts away from the underlying tissue (Figs 5.29 and 5.30).

These lateral folds move down from the cranial end to the caudal end, lifting the whole body away from the underlying tissue (Fig. 5.30). This lifting away from the remaining tissue leaves just one attachment point in the centre, the first evidence of the umbilical cord. The embryo continues to lift off the underlying tissue and the head and tail processes fold back down to give the embryo its characteristic C-shape configuration. At this stage, two more somites are evident, making 16 in total.

The gut tube also now begins to develop from the pharynx fold by closure of the endoderm folds, in a way similar to that by which the neural tube was formed from folds in the ectoderm. The hind-gut of the fetus now extends out into the blastocoel to form the

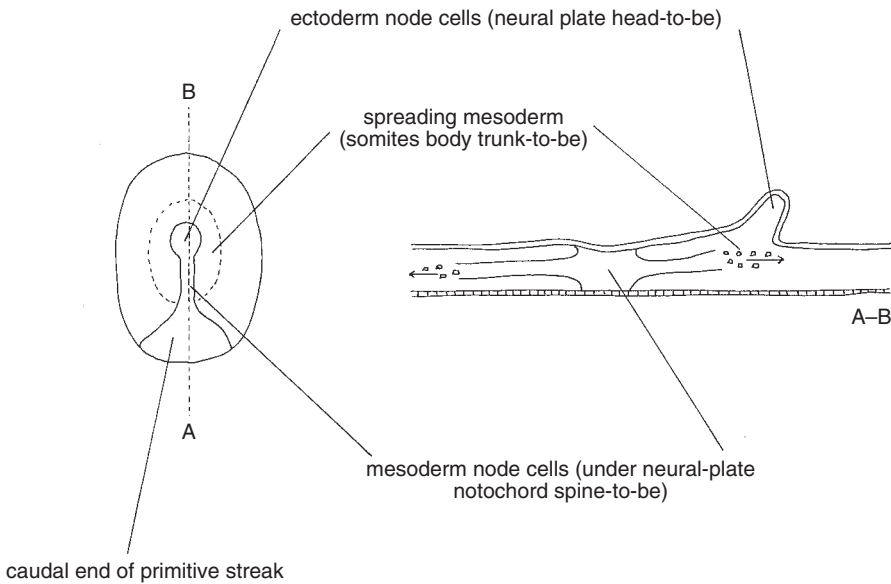


Fig. 5.27. Day 15 completed gastrulation in the equine conceptus. A bird's-eye view and cross-section view through A–B of the embryonic disc. The formation of the head from the cranial end of the neural plate is illustrated, along with the somites, or body trunk, formation from the mesoderm in the immediate vicinity of the neural plate and the spine and central nervous system formation from the mesoderm immediately below the primitive groove.

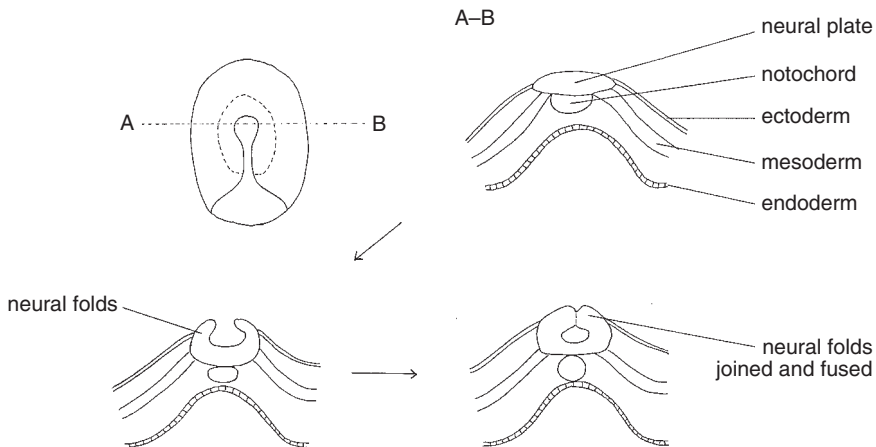


Fig. 5.28. Day 16–17 neurulation of the equine conceptus. The ectoderm near the neural plate thickens and two neural folds develop either side of the neural plate and join to enclose a hollow tube, the future spine and central nervous system.

allantois, as illustrated in Figs 5.10 and 5.11, and blood is also now evident in the lumen of the tubular heart (Cottrill *et al.*, 1997).

The embryo now lies away from the underlying tissue, the placental tissue, and is

connected directly to the mother only by the umbilical cord, which contains a blood system derived from the mesoderm, along with supporting connective tissue. The embryo now has an identifiable neural tube, the forerunner

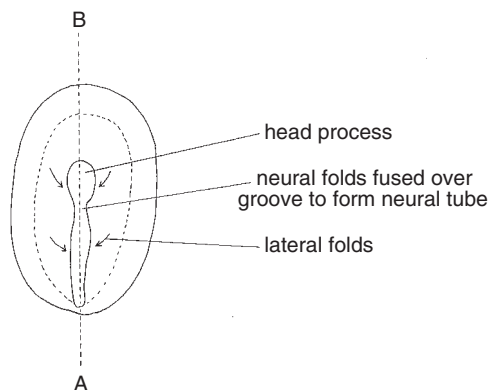


Fig. 5.29. Day 18 neurulation of the equine conceptus. A bird's-eye view of the embryonic shield, illustrating cell movement in towards the cranial end of the neural plate, which subsequently lifts away from the underlying tissue.

of the CNS, and a head process with enlarged neural tube, the brain-to-be. Its pharynx and gut tube are also present, as are the somites, or body muscle blocks. Therefore, by day 23, all the basic bodily structures are evident, though only in a rudimentary form.

5.6. Organ Growth

From day 23 onwards, development is in the form of fine differentiation and organ growth. By day 40, all the main body features are evident, e.g. limbs, tail, nostrils, pig-

mented eyes, ears, elbow and stifle regions, eyelids, etc., and the embryo is now termed a fetus. Day 39–45 heralds sexual differentiation and evidence of external genitalia. The weight of the fetal gonads reaches a maximum at day 180–200, the weight of fetal testis and ovaries being equivalent and developing to the following pattern (Douglas and Ginther, 1975):

Day 80	1.4 g
Day 140	18.7 g
Day 200	48.0 g
Day 320	31.4 g

The increase and decrease in size are due to a proliferation and degeneration of interstitial cells (Walt *et al.*, 1979) and may correspond to the period of masculinization or feminization of the fetus. The reason for the relatively large size of the fetal gonads in the horse is unclear, but it may be related to the secretion of significant levels of oestrogens at this time.

At this stage, most of the development is complete and increase in growth now occurs (Fig. 5.31). At day 60, the eyelids close and finer eye development occurs, teats are present and the palate is fused. Day 160 sees the first evidence of hair around the eyes and muzzle and, by day 180, hair has begun to develop at the tip of the tail and the beginnings of a mane are evident. By day 270, hair covers the whole of the body surface.

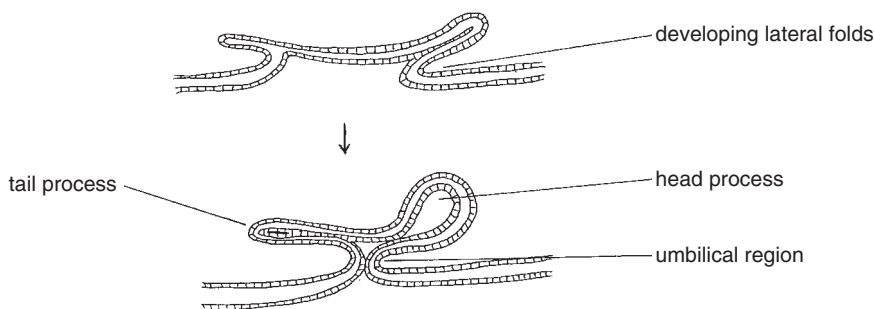


Fig. 5.30. A cross-section view (A–B) of Fig. 5.27, illustrating the gradual appearance of the head and tail processes during neurulation in the 19-day-old equine conceptus. The embryo now begins to take up the characteristic C shape.

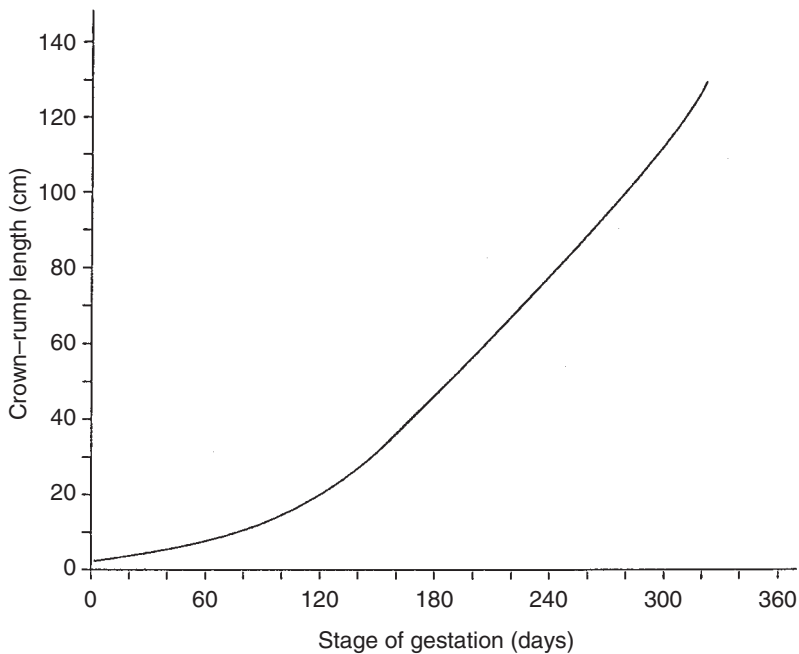


Fig. 5.31. The increase in fetal crown-rump length throughout gestation (data from Roberts, 1971a; Evans and Sack, 1973; Ginther, 1992).

From day 150 onwards, the hippomane, an accumulation of waste minerals within the allantois, becomes apparent. The hippomane increases in size with pregnancy (Fig. 5.32).



Fig. 5.32. The hippomane often found within the placenta of the mare. This example is 11 cm long.

From day 320 onwards, the testes in the male fetus may descend through the inguinal canal; however, this does not occur in all colt fetuses, as some drop neonatally.

The main milestones in equine fetal development are summarized in Table 5.1 (Ginther, 1995; Reef, 1998; Sertich, 1998).

Full term, normally at 320 days in ponies and up to 2 weeks later in Thoroughbred and riding-type horses, heralds the birth of a very well-developed fetus, typical of a preyed-upon, plain-dwelling animal. At birth, foals are capable of all basic bodily functions, including walking, within 30–60 min. Details of the foal's adaptation to the extrauterine environment are given in Chapter 15.

5.7. Conclusion

Our understanding of embryo development specific to the equine is still incomplete, especially in early pregnancy. Continuing

Table 5.1. A summary of the major milestones for fetal development throughout pregnancy.

Day of gestation	Major milestones
1	Zygote, two cells
4	Morula, 16 cells plus
5	Capsule formation
6	Hatching of morula
8	Blastocyst, differentiated into embryonic mass, blastocoel and trophoblast
9	Ectoderm and endoderm germ layers evident, gastrulation begins
11	Segregation giving first embryonic asymmetry, caudal and cranial ends evident
12	Delamination, epiblast cells, hypoblast cells and primitive gut evident
14	Mesoderm evident, primitive streak appearing, involution commencing
15	Primitive streak now evident as a groove
16	Neurulation starts, folds leading to the formation of the amnion seen, first blood vessels evident in mesoderm, chorionic vesicle 2–4 cm diameter
18	Vitelline artery and vein identifiable, fetus begins to take on characteristic C shape
20	Allantois forming from outpushings of the fetal hind-gut, chorionic vesicle oval in shape (2.5–4.5 cm diameter), eye vesicle and ear present
	Capsule begins to degenerate
21	Amnion complete
23	All basic bodily structures evident, though in rudimentary state
25	Chorionic girdle, first evident attachment of fetus
26	Forelimb bud seen, three branchial arches present, eye visible
30	Genital tubercle present, eye lens seen
36	Rudimentary three digits seen on hoof, facial clefts closing, eyes pigmented and acoustic groove forming
40	Endometrial cups forming, ear forming, nostrils seen, eyelids seen, all limbs evident and elbow and stifle-joint areas identifiable, chorionic vesicle 4.5–7.5 cm diameter
42	Ear triangle in shape, mammary buds seen along ridge
45	External genitalia evident, allantoic sac volume 110 ml
47	Palate fused
49	Mammary teats evident
55	Ear covers acoustic groove, eyelids closing
60	Chorionic vesicle 13.3 cm × 8.9 cm
63	Eyelids fused, fine eye development occurring, hoof, sole and frog areas of hoof evident
75	Female clitoris prominent
80	Scrotum clearly seen
90	Endometrial cups degenerate, chorionic vesicle 14 cm × 23 cm
95	Hoof appears yellow in colour
112	Tactile hairs on lips growing
120	Fine hair on muzzle and chin and eyelashes beginning to grow, eye prominent and ergot evident
150	Full attachment of placental microcotyledons, eyelashes clearly seen, enlargement of mammary gland
180	Mane and tail evident
240	Hair of poll, ears, chin, muzzle and throat evident
270	Whole of body covered with fine hair, longer mane and tail hair clearly seen
310	Allantoic-sac volume 8.5 l
320	Testes may drop from this time onwards
320–340	Birth of fully developed fetus

development of our knowledge is essential if we are to understand and hence minimize embryo mortality, a significant cause of apparent infertility in the mare. When the

factors affecting embryo survival are more fully understood, our management of the equine can be further directed towards minimizing losses.

6

Endocrine Control of Pregnancy in the Mare

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6.1. Introduction

When examining the endocrinological control of pregnancy in the mare, gestation can be divided into two stages: early (fertilization to day 150); and late (day 150 to full term).

blocking the drop in progesterone and hence allowing progesterone levels to remain elevated (Fig. 6.1). This is essential for the initial maintenance of pregnancy. If the mare is pregnant, then ovarian progesterone production, from one or a number of corpora lutea (CL), has to be maintained until at least day 75. If there is a failure of the functional CL during this time, then the mare will abort.

6.2. Early Pregnancy (Fig. 6.1)

By day 6, the conceptus has migrated to the uterus and exists by deriving nutrients from the uterine hystotroph or secretions. No major changes from the non-pregnant cycle are evident as yet (Freeman *et al.*, 1991). However, by day 15, a message has to be received by the reproductive system of the mare if it is to continue in pregnancy mode:

6.3. Maternal Recognition of Pregnancy

The importance of day 15 is seen in experiments with early-pregnant mares. If the embryo is removed from a pregnant mare prior to day 15, she will return to oestrus at her normal time (21 days after the last). If,

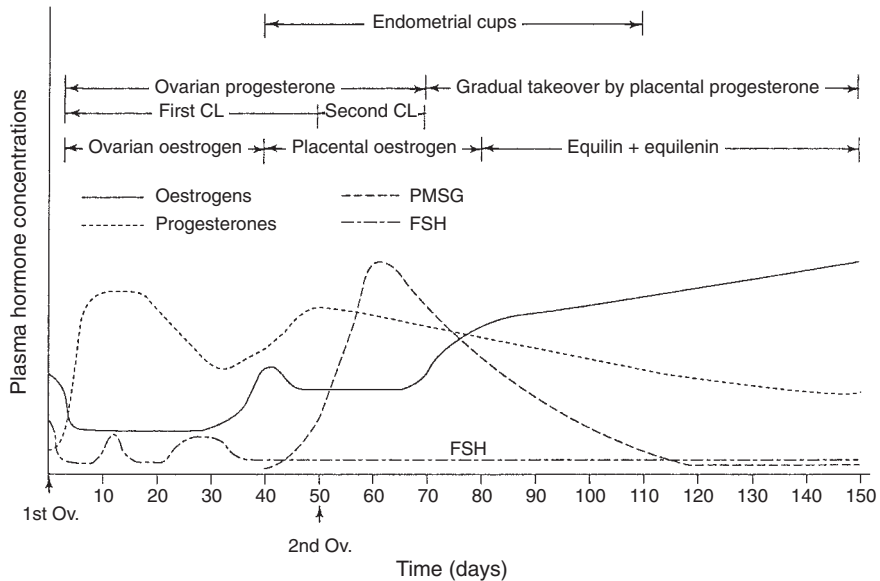


Fig. 6.1. A summary of the plasma hormone concentrations during early pregnancy in the mare (days 0–150). CL, corpora lutea; PMSG, pregnant mare serum gonadotrophin; FSH, follicle-stimulating hormone.

however, the embryo is removed at day 16 or later, then the mare will not return to oestrus as expected and will show a prolonged dioestrus, due to the persistence of the CL. The length of the delay will depend to a certain extent on the age of the embryo at removal.

Experiments show clearly that D-Day as far as the hormonal control of pregnancy is concerned is day 15 (Hershman and Douglas, 1979), though embryo-transfer work suggests there may be a more progressive recognition of pregnancy from day 8 onwards than previously thought (Goff *et al.*, 1987). The exact nature of the message informing the mare of the presence of a conceptus is unclear. However, it has been demonstrated that equine conceptuses are capable of synthesizing oestrogens as early as Day 12 (Flood *et al.*, 1979a; Heap *et al.*, 1982; Sharp, 1993; Choi *et al.*, 1995). As such, oestrogens are the likely candidate acting locally on the uterine epithelium to inform the mother of the presence of the fetus (Flood *et al.*, 1979a; Stewart *et al.*, 1982; Daels *et al.*, 1990). By whatever means the message is delivered, the result is the maintenance of

the CL beyond day 15 (Ball *et al.*, 1991). In the non-pregnant mare, the CL is destroyed at about day 15 by prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), allowing the cyclical changes associated with oestrus and ovulation to begin (Chapter 3). Therefore, in the pregnant mare, this action of $PGF_{2\alpha}$ must be blocked.

The exact mechanism for preventing the action of $PGF_{2\alpha}$ is unknown, but there are several hypotheses. First, it has been suggested that $PGF_{2\alpha}$ binding to the CL is reduced. However, doubt has been placed upon this hypothesis, as it has been reported that the concentration of $PGF_{2\alpha}$ receptors on CL is high during the period days 16–18 postovulation. The second hypothesis is that an alternative component is produced by the uterus, which competitively binds with the $PGF_{2\alpha}$ receptors on the CL. A suitable candidate is prostaglandin E (PGE), which is very similar in structure to $PGF_{2\alpha}$ but biologically inactive with regard to CL regression (Allen, 1970; Heap, 1972; Vernon *et al.*, 1979). Finally the third, and currently favoured, option is that the secretion of $PGF_{2\alpha}$ is reduced. This third hypothesis is supported by: the reported reduction in the concentration of

PGF_{2α} in pregnant-mare uterine washings; the reduction in PGF_{2α} metabolite in the mare's circulation (Kindahl *et al.*, 1982; Zavy *et al.*, 1984); and the ability of the day 12–14 conceptus to suppress the endometrial production of PGF_{2α} *in vitro* (Bazer *et al.*, 1994; Sissener *et al.*, 1996). The equine conceptus is unique in having a relatively long period of mobility (up to 25 days) and it is interesting that, if movement is restricted by ligation, then the greater the restriction, the greater the chance of CL regression (McDowell *et al.*, 1988).

From day 15 onwards, maternal progesterone and fetal oestrogens are the dominant hormones within the uterus and are very important in the production and composition of uterine hystotroph and pregnancy-specific proteins, collectively termed uterine milk. The composition of uterine milk in the mare is very important, as the conceptus survives in a free-living form for a relatively long period of time, no form of implantation occurring in the mare until day 25.

If we consider the hormonal changes individually, then we can build up a picture of how the mare maintains a developing conceptus (see also Squires, 1993e).

6.3.1. Progesterone

Between days 6 and 14 the plasma concentration of progesterone is 8–15 ng ml⁻¹, similar to that seen during dioestrus of the non-pregnant oestrous cycle. Day 15 – decision time – heralds the divergence of progesterone concentrations between the pregnant and non-pregnant mare. Progesterone levels in pregnant mares decline slightly after day 16 (but not to the extent seen in non-pregnant mares), to reach concentrations of approximately 6 ng ml⁻¹ by day 30. Levels subsequently rise again, to reach 8–10 ng ml⁻¹ by day 45–55, and remain at this level, or possibly fall slightly, until day 150 (Holtan *et al.*, 1975b; Schwab, 1990). Experiments in the mare indicate that ovarian progesterone, i.e. that produced by CL, is essential for the maintenance of all pregnancies, at least until day 75 and in some cases

up to day 150. After day 150, placental progesterone is adequate to take over and maintain a pregnancy. All pregnant mares ovariectomized (the ovaries and hence the functional CL removed) before day 75 will abort. If mares are ovariectomized in the period days 75–150, differing reports indicate differing abortion rates. After day 150, ovariectomy has no effect: all mares successfully carrying fetuses to full term (Holtan *et al.*, 1979). This demonstrates that ovarian progesterone is essential prior to day 75 in all mares; in the period days 75–150, placental progesterone gradually takes over and, by day 150, ovarian progesterone is not required. Indeed, at this time, the CL on the ovary can be seen to have regressed.

Ovarian progesterone is not secreted continuously by a single CL. In the mare, an increase in ovarian activity is evident between days 20 and 30 *post coitum*: follicles develop, driven by follicle-stimulating hormone (FSH) surges similar to those seen during dioestrus (Bergfeldt and Ginther, 1992). Dominant follicles become apparent and luteinize between days 40 and 60, forming secondary CL (Chavatte *et al.*, 1997a). These secondary CL are unusual and relatively unique to the mare. During the period days 40–70, the secondary CL gradually take over the production of progesterone, though the primary CL does not necessarily regress. From day 70 onwards, the placenta begins to produce progesterone, gradually taking over the function of all the CL by day 150 (Allen and Hadley, 1974; Evans and Irvine, 1975; Squires and Ginther, 1975; Squires, 1993a).

6.3.2. Equine chorionic gonadotrophin (pregnant mare serum gonadotrophin)

As discussed in Chapter 5, day 40 marks the appearance of endometrial cups. These are responsible for the secretion of equine chorionic gonadotrophin (eCG), also known as pregnant mare serum gonadotrophin. eCG is secreted into the mare's circulatory system and reaches a maximum concentration between days 50 and 70 *post coitum*. It is

secreted by the fetal tissue within the endometrial cups and maximum concentrations achieved vary considerably, different reports giving levels of between 10 and 100 iu ml⁻¹ (Allen and Moor, 1972). Levels are known to be affected by genotype, maximum levels reached and the duration of these levels being greater in mares mated to close relatives, e.g. brother-to-sister mating (Stewart *et al.*, 1977; Allen and Stewart, 1993). The parous state of the mare also has an effect on eCG levels, multiparous having lower levels than primiparous mares. Concentrations always tail off and normally reach basal levels by day 100–120 (Allen, 1982; Allen and Stewart, 1993).

The importance of eCG and why it is only secreted for a short period of pregnancy are unclear. However, several hypotheses suggest its involvement in the prevention of fetal immunological rejection by the mother. An immunological response is also implicated in the reduction in eCG secretion. It appears that the eCG-secreting fetal sides of the endometrial cups are slowly rejected by the maternal system and, as a result, eCG secretion declines until day 100, when all cups have regressed. This hypothesis of an immunological rejection of eCG-secreting fetal cells is supported in work done on sister–brother matings; the fetus from such a mating has a relatively similar genetic make-up to that of its mother. In such pregnancies, eCG is secreted much longer than is seen in non-related matings and endometrial cups regress much later. Presumably, the relatively similar genetic make-up of the fetus does not elicit the same rejection response as in non-related matings.

Linked to this, it has been suggested that eCG is involved in preventing maternal rejection of the developing conceptus. The uterus is a privileged site and is the only place in the body that, under the influence of the hormones of pregnancy, will tolerate a foreign body, the conceptus. The genetic make-up of the conceptus is only half that of its mother and therefore is foreign and, under normal conditions, such an invasion would set up an immunological response and the foreign body (the conceptus) would

be rejected. For some reason, this rejection is prevented in the pregnant mare and all other mammals that carry fetuses *in utero*. It appears that an immunological barrier is set up between the fetus and its mother, blocking the expected immunological response. eCG may be involved in this prevention of rejection from day 40 onwards. However, as it is not secreted prior to day 40, some other component must be responsible for preventing rejection during earlier pregnancy, what this is is unclear (Squires, 1993; Koets, 1995).

eCG has been reported to be involved in follicular development in readiness for the formation of the secondary CL, but recent evidence refutes this (Koets, 1995). eCG is now known not to have FSH-like properties in the mare, though it apparently does in other farm livestock, where it is used as a superovulation agent. If it does have a role in follicle development then this is only via synergizing with natural FSH pulses. eCG does appear to have luteinizing hormone-like properties and is thought to act as a luteinizing agent in the formation of the secondary CL, and it may be implicated as an agent responsible in part for the maintenance of the primary CL (Allen, 1975; Nett *et al.*, 1975; Daels *et al.*, 1991; Allen and Stewart, 1993; Koets, 1995).

6.3.3. Maternal oestrogens

The plasma concentration of maternal oestrogens also varies within pregnancy. Between days 0 and 35, levels remain very similar to those seen during the non-pregnancy dioestrous period, but they then rise sharply to reach 3–5 ng ml⁻¹ by day 40. In the period days 40–45, they decline slightly and subsequently remain at this constant level until day 60–70, after which they slowly rise again (Darenius *et al.*, 1988; Le Blanc, 1991; Stabenfeldt *et al.*, 1991).

These rising levels of maternal oestrogens between days 35 and 40 are thought to be secreted by the developing follicles prior to the formation of the secondary CL, in much the same way as oestrogens are produced by developing follicles prior to ovula-

tion and oestrus in the normal oestrous cycle (Daels *et al.*, 1991). These rising oestrogens, however, do not result in oestrous behaviour. Evidence for the ovarian origin of these oestrogens is their absence in pregnant mares after ovariectomy prior to day 40 and the delayed decline in concentrations seen after fetal death not accompanied by immediate CL regression (Daels *et al.*, 1990, 1991, 1995; Stabenfeldt *et al.*, 1991). However, the second rise in oestrogen at day 60–70 is not affected by ovariectomy but is affected by induced or spontaneous fetal death (Darenius *et al.*, 1988) and can therefore be assumed to originate from the fetoplacental unit. The precursors for such production originate in the fetal gonads (Pashan and Allen, 1979a). By day 85, oestrogens in the mare's peripheral blood system are higher than those detected in non-pregnant mares, and are diagnostic of pregnancy. The continuing rise in oestrogens after day 80 is due to increased fetoplacental production of equilin and equilenin, two oestrogens unique to the pregnant mare (Raeside *et al.*, 1979).

6.4.1. Progesterone

Progesterone levels, which, prior to day 150, were elevated and possibly slowly declining, remain at a steady 1–3 ng ml⁻¹ until day 240–300, after which they steadily decline to basal levels pre-partum. It has previously been reported that progesterone concentrations increase during the last 30–50 days of gestation. However, it is now known that this is not the case, erroneous results arising from the cross-reactivity in old hormone assays between progesterone and its metabolites, in particular 5 α -pregnane, which do increase in the last 30–50 days of gestation (Hamon *et al.*, 1991; Houghton *et al.*, 1991; Squires, 1993a). These high concentrations of progesterone metabolites (progestagens) may themselves have a role pre-partum. They have a similar structure to progesterone but are biologically inactive. As such, high levels of progestagen cause the reproductive system to perceive even lower levels of circulating progesterone, as pregnane molecules attach to the progesterone receptors (Barnes *et al.*, 1975; Moss *et al.*, 1979; Holtan *et al.*, 1991).

6.4. Late Pregnancy

As far as the discussion on hormone control is concerned, late pregnancy can be classified as day 150 onwards (Fig. 6.2).

6.4.2. Oestrogens

Oestrogens within the maternal system continue to rise in late pregnancy, reaching a peak, between days 210 and 280, of approxi-

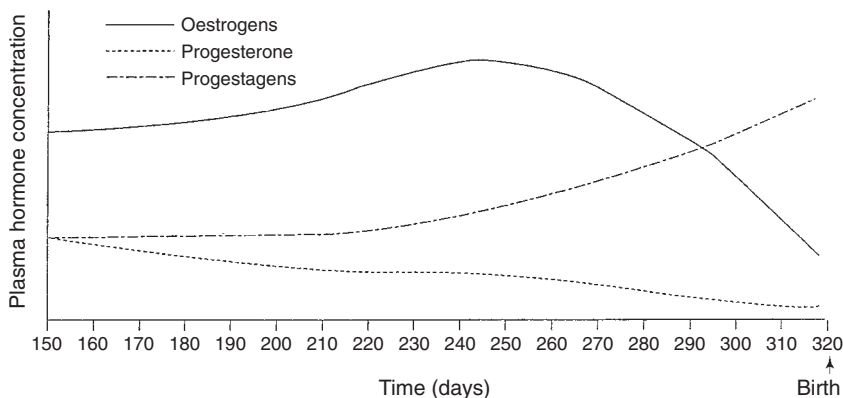


Fig. 6.2. A summary of the plasma hormone concentrations in the mare during late pregnancy (day 150–parturition).

mately 8 ng ml^{-1} , the two main oestrogens being the equine-specific equilin and equilinenin. Oestrogen levels then decline as parturition approaches, reaching levels in the order of 2 ng ml^{-1} at parturition (Raeside *et al.*, 1979; Holtan and Silver, 1992; Le Blanc, 1997).

6.4.3. Prostaglandin $F_{2\alpha}$

During the major part of late pregnancy, $\text{PGF}_{2\alpha}$ remains at low levels, equivalent to those seen during early pregnancy. However, near to term, levels do increase slightly, in the form of short pulses, but significant elevations in concentration are not detected

until labour has started, when they play a major role in uterine myometrial contraction (Barnes *et al.*, 1978).

6.5. Conclusion

It is evident that the control of pregnancy in the mare follows a similar but not identical pattern to that seen in other mammals, with a dominant role played by progesterone and oestrogen throughout. Additionally and uniquely to the mare, eCG is produced and is involved in the formation and maintenance of secondary CL, which are required to maintain the dominance of progesterone.

7

The Physical Process of Parturition

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7.1. Introduction

Parturition is the active expulsion of the fetus, along with its associated fluid and placental membranes. The average gestation length in the mare is 320–335 days. It tends to be up to 2 weeks shorter in ponies than in Thoroughbreds and horses of the larger riding type (Rophia *et al.*, 1969; Rosedale *et al.*, 1984).

There are several signs that indicate that parturition is approaching. These may become evident at any time in the last 3 weeks of pregnancy. It must be remembered that these signs, detailed below, should not be used in isolation and that there is much variation between individuals and between successive pregnancies. Therefore, a combination of the following signs should be looked for when watching for imminent parturition. It is also useful to have information on a mare's previous pregnancies, as general behavioural patterns may be characteristic to a particular mare.

7.2. Signs of Imminent Parturition

Changes in the appearance of the udder are one of the first signs of imminent parturition. During the last month of gestation, as lactogenesis (milk production) commences, the udder increases in size as colostrum is produced and stored (Forsyth *et al.*, 1975; Peaker *et al.*, 1979). The udder may feel relatively warm to the touch, as a result of the increased cell metabolic activity associated with milk production. At this time, the udder may seem to increase in size at night, especially if the mare is kept in, and decrease during the day when she is turned out and able to exercise, exercise being associated with increased circulation and reduction in udder oedema (fluid accumulation). When there is no apparent change in udder size between day and night, parturition is imminent. At this stage, the udder is so full of milk that exercise no

longer affects its size. The extent to which udder size increases is dependent upon the size of the mare and her parity (number of previous foals) (Rossdale and Ricketts, 1980). Figure 7.1 shows the udder of a Welsh Cob Section D mare 5 days prior to parturition.

The teats also change, initially becoming shorter and fatter as the udder fills and the bases of the teats are stretched. As the time for parturition approaches, the teats fill as milk production increases; they elongate and become tender to touch. Some mares may even start to lose milk as the production by the udder gets too great for its storage capacity and the rosette of Fustenburg sphincter at the end of the teat is breached. If a mare does start to lose milk, it is very important to minimize the loss. The milk at this stage is in fact colostrum, with a high concentration of immunoglobulins, and is responsible for the transfer of passive immunity to the foal. As there is a finite amount of colostrum, if a mare is seen to lose milk or habitually does so, it is a good idea to milk her out a bit and store the collected colostrum for feeding to the foal immediately post-partum. Colostrum can be successfully frozen for more than a year for use at a later date (Peaker *et al.*, 1979).

Many mares 'wax up', a term given to the clotting of colostrum at the end of the teat (Fig. 7.2). This is a good sign of imminent parturition. However, the lack of wax is not indicative that parturition is not imminent, as these plugs of colostrum can easily be dislodged, especially in active mares.

The concentration of several minerals – sodium, phosphorus (P), calcium (Ca) and potassium (K) – within the mammary gland secretions as parturition approaches is also indicative of the imminence of parturition. These parameters are advocated for use when attempting to assess fetal maturity prior to the artificial induction of parturition (see Chapter 15; Ousey *et al.*, 1984). In particular, Ca concentrations can be assessed via water-hardener strips (Ley *et al.*, 1993). Ca concentrations in excess of 10 mmol l^{-1} are reported to be evident immediately prior to parturition (Ousey *et al.*, 1984; Cash *et al.*, 1985; Brook, 1987).

Changes in the birth canal also become apparent as parturition approaches. Approximately 3 weeks prior to parturition, hollow-ness may appear either side of the tail root as the muscles and ligaments within the pelvic area relax. The whole area may appear to sink with this relaxation and so allow expansion of the birth canal during the passage of



Fig. 7.1. The udder of a Section D mare 5 days prior to parturition.



Fig. 7.2. One of the signs of imminent parturition is the accumulation of dried colostrum on the teats of the mare, termed waxing up; dried colostrum may also be seen on the insides of the hind legs.

the foal. If the area either side of the tail root is felt daily in the last 3–4 weeks of pregnancy, it may be possible to detect a change as the muscle tone relaxes (Fig. 7.3).

Changes in the mare's abdomen may also be evident in late pregnancy. As the fetus increases in size, the abdomen expands correspondingly, becoming characteristically large and pendulous. However, in the final stages of pregnancy, the abdomen appears to shrink as the foal moves up out of the

lower abdomen and into the birth canal ready for delivery (Fig. 7.4).



Fig. 7.3. A further sign of imminent parturition is a hollowing of the back above the pelvis, as a result of a relaxation of the birth canal.

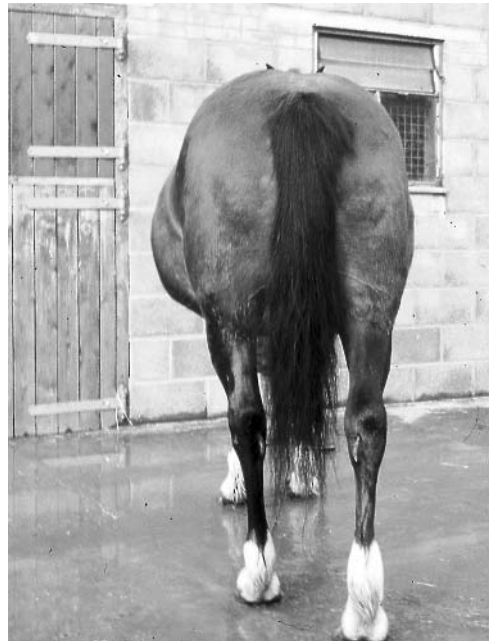


Fig. 7.4. One of the obvious signs of pregnancy is a large pendulous abdomen. However, immediately prior to parturition, the size of the abdomen appears to shrink as the foal moves up into the birth canal.

As parturition approaches closer still, the mare becomes restless and agitated, especially as she enters first-stage labour. Some restlessness may also be apparent in late gestation, and naturally, at this stage, the mare would move away to the periphery of the herd in readiness to move away completely for labour itself. As the mare moves into first-stage labour, her body temperature increases and she may sweat profusely (Haluska and Wilkins, 1989). Internally, her cervix will dilate (Volkman *et al.*, 1995). During first-stage labour, she may show signs very similar to those indicative of colic, e.g. walking in circles, swishing tail, looking around at her sides, kicking her abdomen, etc. If a mare does show signs of colic in late gestation, it is pertinent to consider that it may in fact be first-stage labour, and so her eating, drinking and defecating should be checked.

As discussed, not all mares show all these symptoms, but a combination of one or two will give an accurate prediction that foaling is imminent. Some commercial products have been produced to aid in the diagnosis. These make use of some of the mare's natural signals, such as the mare's movements, increase in body temperature or sweating, stretching of the vulval lips, etc. (Shaw *et al.*, 1988; Amann *et al.*, 1989). Once triggered, they normally produce a signal transmitted to an audio receiver. Closed-circuit television is also popular, allowing discrete observation.

7.3. The Process of Parturition

Parturition in most mammals involves three distinct stages: stage 1, positioning of the foal and preparation of the internal structures for delivery; stage 2, the actual birth of the foal; and stage 3, the expulsion of the allanto-chorion and amnion (placental membranes). All three stages involve considerable myometrial activity, mainly within the uterus itself, but with some involvement of the abdominal muscles (Card and Hillman, 1993).

7.3.1. First stage of labour

Stage 1 involves uterine myometrial contractions and lasts between 1 and 4 h, positioning the foal ready for birth. Figure 7.5 illustrates the forces involved in this stage (Ginther, 1993).

The uterine muscles contract in mild waves from the tip of the uterine horn towards the cervix. These contractions, helped by the movement of the mare and, to a certain extent, by those of the foal, result in the repositioning of the foal and its passage into the birth canal, the area of least resistance (Haluska *et al.*, 1987a,b). Throughout late pregnancy the foal lies in a ventral-flexed position (its vertebrae lying along the line of the mother's abdomen) with its forelimbs flexed. As first-stage

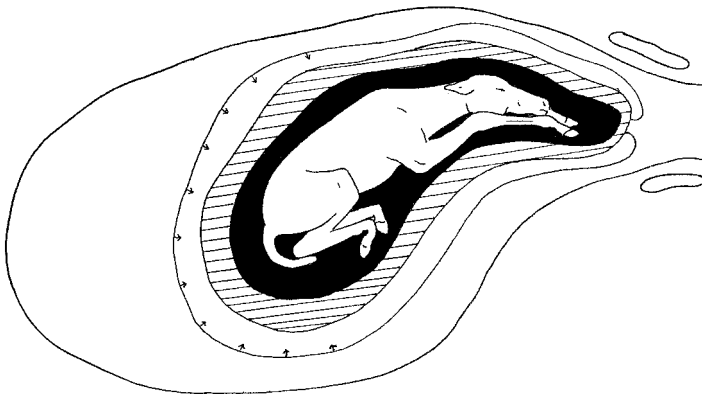


Fig. 7.5. The forces involved in the first stage of labour are provided by contractions of the uterine myometrium, as indicated by the arrows.

labour approaches and during first stage, it rotates into a dorsal position, with its forelimbs, head and neck fully extended, becoming engaged in the birth canal (Fig. 7.6). This movement occurs normally 3–4 h prior to the commencement of labour. In some cases, a mare may show signs of first-stage labour and then cool off, only to show further signs several hours later; this is particularly evident in Thoroughbred mares (Jeffcote and Rossdale, 1979; Rossdale and Ricketts, 1980).

If parturition is to continue, other changes, in addition to the engaging of the foal, must occur. The cervix gradually

dilates. This is encouraged during the later part of first-stage labour by the pressure of the allanto-chorionic membrane and the foal's forelimbs against the uterine side of the cervix. During the birth of a dead fetus, dilatation of the cervix is less complete and slower, presumably as cervical dilatation is actively encouraged by the movements of the foal (Volkman *et al.*, 1995).

The vulva continues to relax and secretions increasingly collect within the vagina. The exact mechanism by which parturition myometrial activity is induced and controlled is unclear (see Chapter 8). However, active movement of the foal against the

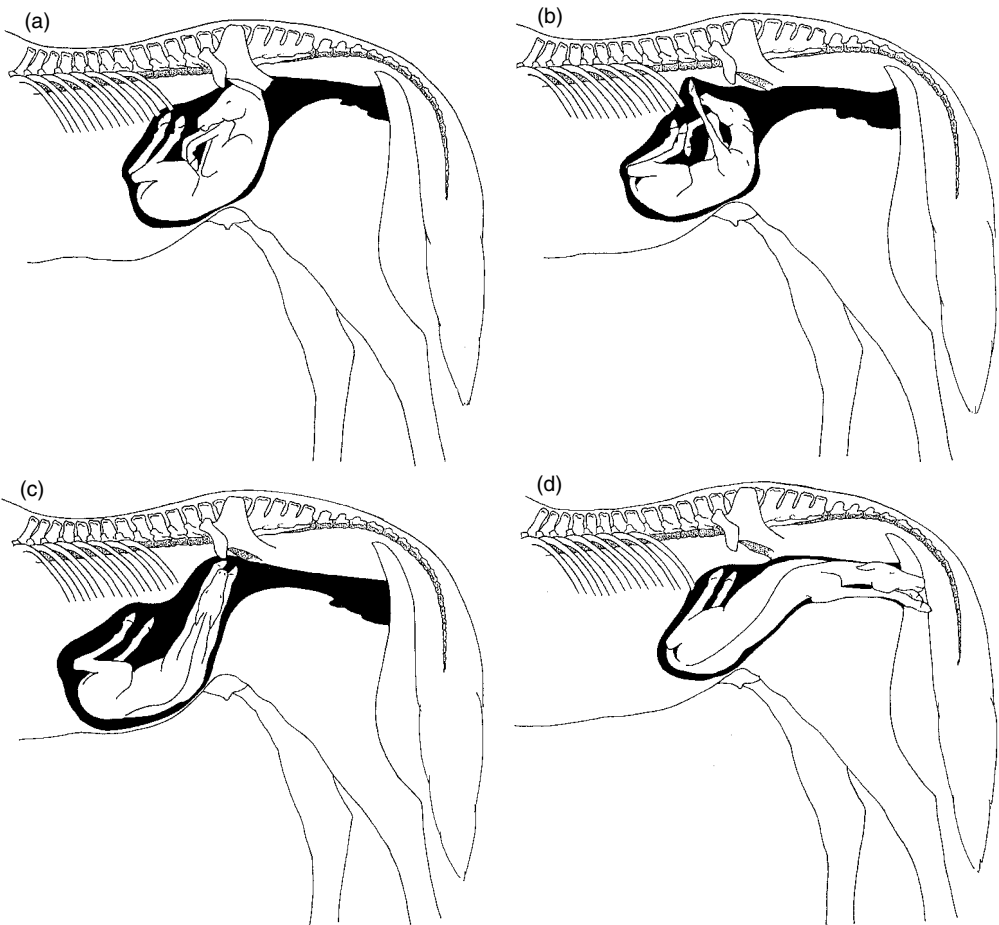


Fig. 7.6. During the first stage of labour the foal is gradually rotated and positioned within the birth canal in readiness for expulsion during the second-stage contractions.

cervix and within the birth canal has been shown to increase prostaglandin F metabolite, and hence, by inference, prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) levels. Such an increase in $PGF_{2\alpha}$ is not seen during the birth of a dead foal. The cervix is high in collagen. The ratio of collagen to muscle fibres progressively increases from the uterine horn through the uterus to the cervix. It is thought that $PGF_{2\alpha}$ affects this collagen, causing it to relax. A similar effect has been shown in the ewe (Volkman *et al.*, 1995). The hormone relaxin may also have a role (Bryant-Greenwood, 1982).

At the end of first-stage labour, the fetus's forelegs and muzzle push their way through the dilating cervix, taking with them the allanto-chorion. At the cervix, this membrane, termed the cervical star, is one of the three sites devoid of microcotyledons and therefore there is no attachment to the maternal endometrium. The other two are at the entry to each Fallopian tube. The cervical star is the thinnest area of the placenta and hence it is this area that ruptures as the pressure of the myometrial contractions against the placental fluids increases, forcing them and the fetus through the cervix. The subsequent release of the allantoic fluid (breaking of the waters) is the signal for the beginning of the second stage of labour.

7.3.2. Second stage of labour

The release of allantoic fluid at the start of stage 2 lubricates the vagina and is thought to trigger the stronger uterine contractions of the second stage. These strong contractions continue until the birth of the foal, normally within 20 min (acceptable range 5–60 min) (Rossdale and Ricketts, 1980). At the start of stage 2, the amniotic sac is often visible bulging through the vulva (Fig. 7.7), within which should be felt the foal's forelegs and muzzle (Figs 7.8 and 7.9). Second-stage labour involves stronger contractions of the uterine myometrium, supplemented by abdominal-muscle contractions.



Fig. 7.7. The amniotic sac should be seen as a white membrane protruding from the mare's vulva (photograph Steve Rufus).



Fig. 7.8. At the start of second-stage labour, a brief internal examination may be made to ascertain whether or not the foal is correctly presented (photograph Steve Rufus).



Fig. 7.9. If all is well, the mare should be left to foal unaided and observed from a discreet distance (photograph Steve Rufus).

The supplementary force provided by the abdominal muscles is termed voluntary straining. During voluntary straining, the mare inspires deeply, holding the rib cage and diaphragm at maximum inspiration. The rib cage and abdominal muscles react by contracting against the pressure; this additional contraction force is transferred to the uterine contents, adding extra impetus to the expulsion of the uterine contents (Fig. 7.10).

At the end of first-stage labour, the foal lies in a dorsal extended position. The soft tissue and bones surrounding the birth canal govern its shape. The pelvis contains the sides and bottom of the canal and the sacral and coccygeal vertebrae the top. The diameter of the entry into the birth canal (20–24 cm) is slightly larger than the exit diameter (15–20 cm) and slightly more dorsal (nearer the mare's vertebrae) than the exit. Hence, the foal is funnelled through the birth canal in a curved manner, being expelled ventrally (down towards the mare's hind legs) (Figs 7.11 and 7.12) (Rossdale and Ricketts, 1980).

The foal is delivered forelegs first, followed by the head lying extended between the forelegs, parallel with the knees. The two forelegs are not delivered aligned: normally one leg is delivered slightly in advance of the other, the fetlock of one being in line with the hoof of the other (Fig. 7.13). This misalignment of the forelegs reduces the cross-section diameter of the foal's thorax, which is the widest part of the foal, so reducing foal trauma at birth and easing foaling. Once the thorax and shoulders have passed through the birth canal, the remainder of the birth process is relatively easy (Rossdale and Ricketts, 1980). At the end of stage 2, the foal lies with its head near the mare's hind legs, with its own hind limbs still within the mare (Fig. 7.12). The presence of the foal's

Fig. 7.10. Second-stage labour involves stronger contractions of the uterine myometrium, supplemented by contractions of the abdominal muscles, as indicated by the arrows.



Fig. 7.11. The angle of the birth canal dictates that the foal is delivered in a curved manner, being expelled down towards the mare's hind legs (photograph Steve Rufus).



Fig. 7.12. The end of the second stage of labour is marked by the foal lying with its hind legs still within the vulva of its mother (photograph Steve Rufus).

legs within the mare's vagina has an apparent tranquillizing effect, most mares being reluctant to rise immediately post stage 2 (Rossdale and Ricketts, 1980).

7.3.3. Third stage of labour

Stage 3 of labour is normally completed within 3 h of the end of stage 2. Uterine contractions continue at a level similar to that evident during stage 1 labour, again originating at the uterine horns and passing down in waves to the cervix. At the same time, the allanto-chorion begins to

shrink, as blood is drawn away from it towards the foal's pulmonary system. The blood vessels constrict and draw the allanto-chorion away from the uterine endometrium. This releases the remaining attachments between the allanto-chorion and the uterine epithelium and forces the placenta to be expelled inside out. The placenta is delivered with its red, velvety, outer allanto-chorion innermost and the smooth, inner allanto-chorion outermost (Figs 7.14 and 7.15). The contractions of third-stage labour also help to expel any remaining fluids and assist in uterine involution (recovery).



Fig. 7.13. In order to reduce the diameter of the foal's thorax passing through the birth canal, the foal is delivered with one foot slightly in advance of the other (photograph Steve Rufus).

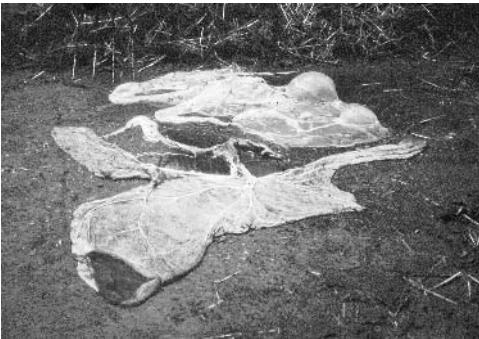


Fig. 7.14. The amniotic sac, the white membrane in the background, and the allanto-chorion (placenta proper), in the foreground, after expulsion during the third stage of labour. As can be seen, the placenta is expelled inside out, the red, velvety, outer allanto-chorion being inside and the smooth inner allanto-chorion being the outermost.

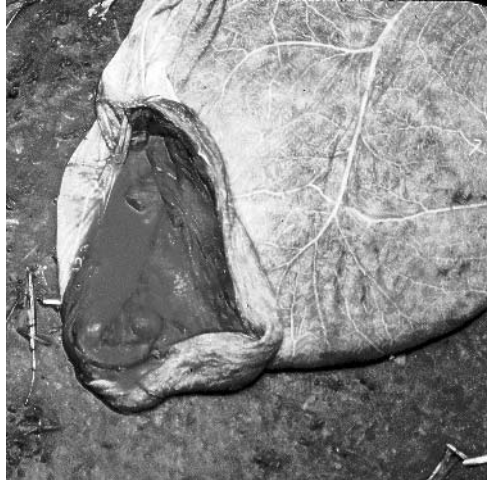


Fig. 7.15. A close-up of the placenta in its inside out state, illustrating the innermost red allanto-chorion and the outermost smooth white allanto-chorion.

7.4. The Hippomane

Passed out either during second-stage labour or along with the placenta during third-stage labour is a small, brown, leathery structure termed the hippomane (Fig. 5.32). It is found within the allantoic fluid and is an accumulation of waste salts and minerals collected throughout pregnancy. It has a high concentration of calcium, magnesium, nitrogen, phosphorus and potassium, and is first evident at around day 85 of pregnancy. It has been associated with much folklore, including aphrodisiac properties and responsibility for keeping the foal's mouth open. There is, however, no evidence to support these claims.

7.5. Conclusion

The diagnosis of imminent parturition is challenging in the mare, which may demonstrate a range of signs over a period of time. However, once started, parturition itself occurs rapidly over a matter of an hour or so and without problems, requiring minimal interference from humans.

8

The Endocrine Control of Parturition

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8.1. Introduction

Parturition in the mare occurs at approximately 11 months (310–374 days) after conception. The exact length of gestation, however, varies with the type of horse. Ponies tend to foal on average 2 weeks before Thoroughbred mares, at about 320 days compared with 335 days (Rophia *et al.*, 1969; Rosedale *et al.*, 1984).

Within these averages, many factors may influence the exact timing of parturition, including environmental, fetal and maternal factors. Environmental factors include the season of mating. Mares mated early in the season tend to have longer gestations than those mated later on. This is presumably nature's way of compensating for early and late matings, trying to ensure that all mares

foal down at the optimum time of the year for foal survival – that is, during spring (Howell and Rowlin, 1951; Hodge *et al.*, 1982). Climate (Astudillo *et al.*, 1960), year of breeding or foaling (Howell and Rowlin, 1951; Rophia *et al.*, 1969) and nutrition are also reported to have an effect. Lastly, nutritional deprivation in the final trimester is reported to cause early parturition.

Fetal factors include the genotype of the offspring. This can be demonstrated by comparing the gestation lengths of various crosses within the equine species. A stallion-cross-mare fetus has an average gestation of 340 days; for a stallion-cross-jennet fetus, it is 350 days, a jack-cross-mare fetus 355 days, and a jack-cross-jennet 365 days (Rollins and Howell, 1951; Ginther, 1992). Breed of foal is also a reported factor (Jochle, 1957), as is foal

gender; colt foals have gestations on average 2.5 days longer than filly foals (Rophia *et al.*, 1969; Hevia *et al.*, 1994; Panchal *et al.*, 1995). Multiple births also have shorter gestations than singles (Jeffcote and Whitwell, 1973).

Finally, we come to maternal factors: the mare herself has some control over the exact time of delivery. The majority of mares foal at night and undisturbed (Fig. 8.1; Rossdale and Short, 1967; Goater *et al.*, 1981; Roberts, 1986c; Hines *et al.*, 1987). There is evidence that this is linked to a circadian rhythm in oxytocin secretion around parturition (Nathanielsz *et al.*, 1997). In addition, the mare's age (Bos and Van der May, 1980), parity (Britton and Howell, 1943; Panchal *et al.*, 1995), foaling-to-conception interval (Britton and Howell, 1943), genotype (Rophia *et al.*, 1969) and breeding-to-ovulation interval (Ganowiczowa and Ganowicz, 1966) have all been reported to affect gestation length.

8.2. Initiation of Parturition

Birth involves the expulsion of the fetus plus all associated placental membranes and fluid and is achieved by myometrial activity within the uterus and surrounding structures (see Chapter 7). Myometrial activity is inhibited by elevated progesterone and depressed oestrogen concentrations, characteristics of pregnancy in most mammals. At full term, the ratio of progesterone to oestrogen reverses, removing any inhibition and allowing myometrial activity to be driven by elevated prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) and oxytocin concentrations, both of which can be seen to rise at parturition. Efficient expulsion of the fetus and placental tissue is dependent upon sequential contraction of the whole uterine myometrium. There must, therefore, be immediate activation of muscle cells and efficient cell-to-cell induction. This message transfer is affected by circulating hormone concentrations. Elevated proges-

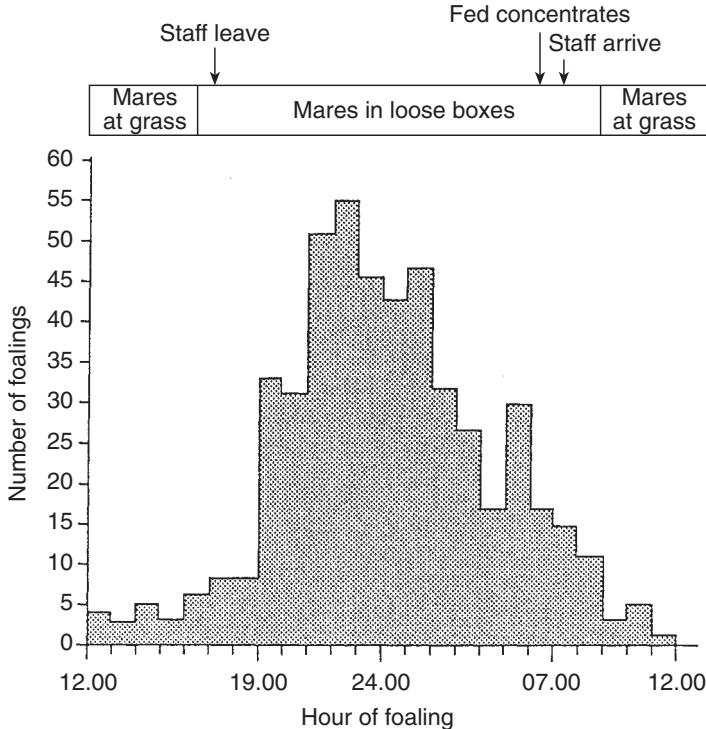


Fig. 8.1. The time of foaling in the Thoroughbred mare (from Rossdale and Short, 1967).

terone concentrations reduce the spread of muscle-cell excitation and contraction (Liggins, 1979; Holtan *et al.*, 1991; Rossdale *et al.*, 1997). The exact mechanism for the initiation of this myometrial contraction of parturition in the horse or any other Equidae is as yet unclear. However, in other mammals, two alternative mechanisms are apparent.

First, as seen in the ewe, nanny-goat, sow and cow, the fetus itself actively controls the initiation of its own parturition. Towards the end of gestation, the fetus comes under considerable stress, due to hypoxia (a shortage of oxygen), its physical restriction within the uterus and a shortage of nutrient supply. These increasing stress levels activate the fetal hypothalamic-pituitary-adrenal axis, causing the production of fetal corticosteroids. These pass to the placenta and change the metabolic pathways, so converting progesterone to oestrogen (Power and Challis, 1987). As a result, the characteristic rise in oestrogens and fall in progesterone near term in these animals is achieved.

The second apparent method by which parturition is initiated is seen in primates. In such mammals, the start of parturition is controlled by a genetically controlled maturation signal linked closely to the time of gestation. As a result, the fetal adrenals produce increasing amounts of androgens, the precursor for placental oestrogens, and no involvement of the fetal adrenocorticoids is evident. However, the result is the same: increasing oestrogen concentrations.

Regardless of the exact initiation of parturition in both hypotheses, the end result is an increase in the ratio of oestrogen to progesterone. Oestrogen is central to the initiation of the strong contractions of first- and second-stage labour (Nathanielsz *et al.*, 1997). The link between increasing oestrogen and increasing oxytocin production and oxytocin receptors on the endometrium is well documented (Fuchs *et al.*, 1983; Wu and Nathanielsz, 1994). The central role of oxytocin in parturition is increasingly becoming evident and it is possibly more central than $\text{PGF}_{2\alpha}$ (Nathanielsz *et al.*, 1997). $\text{PGF}_{2\alpha}$ production by the endometrium is also linked to increasing oestrogen levels. So, as oestrogen

production by the placenta increases, oxytocin and $\text{PGF}_{2\alpha}$ concentrations rise. Both these are major activators of uterine myometrial activity. Additionally, there is evidence of a circadian rhythm in oxytocin production. This rhythm is inherent but may be modulated by daylight, night-time being associated with increased oxytocin production (Nathanielsz *et al.*, 1997), so in part explaining the increased incidence of parturition during the hours of darkness. The mare herself also appears to have some fine control over the exact timing of oxytocin release.

In summary, once the inhibition of progesterone is removed, elevated oestrogen levels drive the production of oxytocin and $\text{PGF}_{2\alpha}$, which in turn drive the uterine myometrial activity associated with contractions.

Though there is no conclusive evidence as to which mechanism occurs in the mare, most of the evidence available indicates a system similar to that seen in the ewe (Silver, 1990; Silver and Fowden, 1994). Prolonged elevated corticosteroid levels in the later part of gestation have not been reported in the equine fetus, though this may be due to difficulties encountered in catheterizing such fetuses. However, significantly elevated corticosteroid levels have been reported in the equine fetus immediately prior to parturition, reaching a peak 30–60 min post-partum (Silver and Fowden, 1994). It has been demonstrated that cortisol is essential in foals for the final maturation of several internal organs, including the respiratory and digestive tract. Pashan and Allen (1979a,b) presented evidence that parturition in the equine is influenced by fetal stress, via an interaction between fetal and placental size, and that fetal constriction may be a trigger. This hypothesis is further supported by Rossdale *et al.* (1992), who demonstrated that treatment of fetuses *in utero* with adrenocorticotrophic hormone (ACTH) resulted in an increase in corticosteroid production by the fetal adrenals, which caused premature parturition, as observed in ewes. Cortisol levels immediately prior to parturition are elevated more dramatically but over a shorter period of time than is evident in the ewe and cow. The relatively short period of

elevated cortisol levels apparently reflects the rapid maturation of the equine fetal adrenals, which is a necessity for post-partum survival and which occurs in the last 2–3 days of gestation (Chavatte *et al.*, 1997b; Le Blanc, 1997; Rossdale *et al.*, 1997).

8.3. Endocrine Concentrations

The hormones involved in parturition will be considered in turn (Fig. 8.2).

8.3.1. Oestrogen

Oestrogen concentrations in the maternal blood system fall over the last 30 days of gestation and, as they originate from the fetal–placental unit, reach basal levels within hours of parturition. Concentrations are approximately 6 ng ml^{-1} at 30 days prior to parturition and fall to less than 2 ng ml^{-1} after parturition (Nett *et al.*, 1973).

8.3.2. Progesterone

Progesterone concentrations decline to basal levels at parturition, though concentrations of progesterone metabolites, such as 5α -pregnanones, peak at $10\text{--}15 \text{ ng ml}^{-1}$ 10 days pre-partum (Hamon *et al.*, 1991; Holtan *et al.*, 1991; Schutzer and Holtan, 1995). These metabolite concentrations reflect an alteration in placental metabolism towards oestrogen production during late pregnancy and parturition. These progestagens also occupy the progesterone binding sites within the uterine endometrium and, as such, their increase in concentration is perceived by the uterus as a further decrease in circulating progesterone. It is likely that it is the ratio of oestrogen to progesterone, rather than the absolute levels, which is important. Therefore, even though oestrogen levels decline, it is possible that their decline is significantly less than that perceived for progesterone, with, in relative terms, the ratio of oestrogen to progesterone increasing, allowing myometrial activity to commence (Purvis, 1972; Moss *et al.*, 1979; Rossdale *et al.*, 1991).

8.3.3. Prostaglandin $F_{2\alpha}$

As discussed previously (Chapter 2), prostaglandin $F_{2\alpha}$ metabolite (PGFM) is used as an indicator of $\text{PGF}_{2\alpha}$ concentrations. From measurements of PGFM, it can be deduced that $\text{PGF}_{2\alpha}$ concentrations rise sharply in the peripheral plasma of the mare at term, mainly during the second stage of labour. Fetal $\text{PGF}_{2\alpha}$ levels, as determined by catheterization, increase more gradually over the final weeks (40 days) of pregnancy (Barnes *et al.*, 1978; Cooper, 1979; Silver *et al.*, 1979). $\text{PGF}_{2\alpha}$ may also be detected in the allantoic fluid near to parturition. The major functions of $\text{PGF}_{2\alpha}$ are to induce cervical dilatation and as a strong inducer of uterine myometrial activity. $\text{PGF}_{2\alpha}$ is primarily associated with first- and second-stage labour.

8.3.4. Oxytocin

The actions of $\text{PGF}_{2\alpha}$ and oxytocin are closely linked and they tend to show the same pattern of release. Oxytocin concentrations in the maternal system rise sharply at parturition, especially during the second stage. The central role of oxytocin is increasingly becoming evident and, like $\text{PGF}_{2\alpha}$, its primary role is in the induction of strong myometrial contractions, in particular those of second-stage labour, when it works in concert with $\text{PGF}_{2\alpha}$ (Fuchs *et al.*, 1983; Haluska and Currie, 1988). Oxytocin is also involved in the myometrial contractions of the third stage of labour (Hillman and Ganjam, 1979), unlike $\text{PGF}_{2\alpha}$, which is primarily involved in the first and second stages. The reported circadian rhythm of oxytocin release has been discussed previously.

8.3.5. Cortisol

Cortisol concentrations do not change significantly in the maternal system during pregnancy, though they rise, due to stress, at parturition. Changes within the fetal sys-

tem occur in late pregnancy, with a sudden rise in cortisol evident just prior to parturition (Nathanielsz *et al.*, 1975; Liggins *et al.*, 1979). As previously discussed, this increase is shorter and sharper than the gradual increase observed in the sheep and goat and is known to be associated with the maturation of the fetal adrenal cortex and its ability to react to circulating ACTH (Silver and Fowden, 1994; Nathanielsz *et al.*, 1997). It may well also be involved in final organ maturation – for example, the respiratory system – in the equine fetus (Rossdale *et al.*, 1973; Alm *et al.*, 1975).

8.3.6. Prolactin

Prolactin concentrations are reported to increase in the last 7–10 days. It is not apparent that prolactin has a direct role in parturition, but its increase at this time may indicate a role in equine lactation, as seen in other mammals (Forsyth *et al.*, 1975; Worthy *et al.*, 1986; Nett, 1993b).

8.3.7. Relaxin

Plasma concentrations of relaxin are reported to be elevated in late pregnancy/parturition, its production site being the placenta. In late pregnancy, it is thought to maintain the quiescent nature of the uterine myometrium, but, during parturition, it is overcome by the activation of oxytocin and $\text{PGF}_{2\alpha}$. Relaxin may also have a role in the relaxation and softening of the pelvic ligaments and cervix as parturition approaches (Bryant-Greenwood, 1982).

8.4. Conclusion

It is evident that the mechanism for the initiation of parturition in the mare is unclear, but seems probably to be due, at least in part, to fetal stress. Confirmation of this hypothesis is largely hampered by the limitation of experimental techniques to date. Clarification of the hormonal control of equine parturition would enable more accurate diagnosis of imminent parturition and artificial induction of parturition.

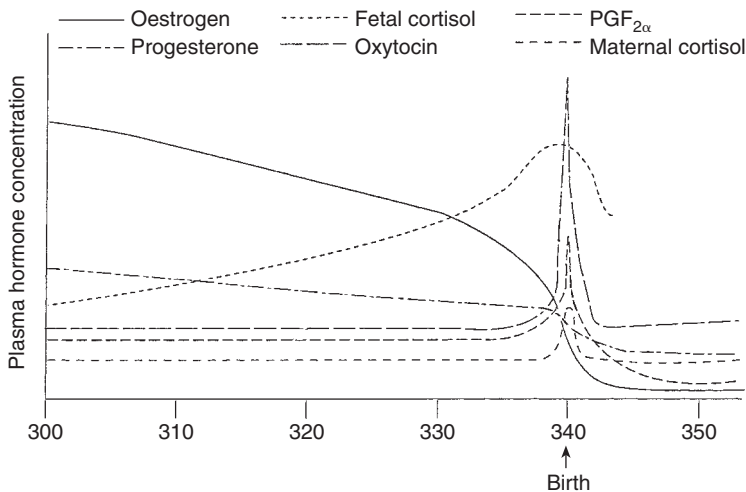


Fig. 8.2. A summary of the main changes in hormone concentration evident prior to parturition. $\text{PGF}_{2\alpha}$, prostaglandin $\text{F}_{2\alpha}$.

9

The Anatomy and Physiology of Lactation

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9.1. Introduction

The mammary glands of the mare, in common with all mammals, are modified apocrine sweat glands, both developing *in utero* from a common precursor. They are situated along the ventral midline in all mammals in a varying number of pairs. The mare normally has four glands – two pairs.

In most mammals, each gland exits via its own teat; however, in the mare each pair of glands on either side of the midline exits via a single teat. The anatomy and physiology of lactation specific to the equine have not been widely studied. Other reviews on the subject include Jacobson and McGillard (1984), Mephram (1987), Kainer (1993) and McCue (1993).

9.2. Anatomy

The mammary glands of the mare are situated in the inguinal region between the hind legs, covered and protected by skin and hair, except for the teats, which are devoid of hair. The whole of the skin surface is supplied with nerve endings, the concentrations of which are increased in the teat area, enhancing the response to touch. The mare normally has four glands, two larger cranial ones and two smaller caudal ones, though six glands have been reported in the occasional mare. Each of the four mammary glands is completely independent, with no passage of milk from one quarter to another. They are separated by and contained within a fibroelastic capsule and supported by the medial suspensory ligament, running along the mare's midline. Further support is provided by the lateral suspensory ligaments, running over the surface of the mammary gland itself under the skin, and by laminae, developing off the suspensory ligament and penetrating the mammary tissue in sheets (Fig. 9.1; Sisson, 1975; Kainer, 1993).

Each udder half, either side of the midline, is made up of two quarters and the openings from these two quarters exteriorize via a single teat (Fig. 9.2).

The mammary tissue itself is made up of millions of alveoli and connecting ducts. This arrangement can be compared to a bunch of grapes, each alveolus being equated to a grape and the ducts to the branches (Fig. 9.3).

The alveoli are grouped together in lobules and then the lobules into lobes. These lobes join together via a branching duct system, which eventually leads to the gland cistern. Each quarter has its own gland cistern draining into a teat cistern and on to the streak canal, one from each quarter on that side. At the end of each teat is the rosette of Fustenburg, a tight sphincter that prevents the leakage of milk between sucklings. This sphincter can withstand a considerable build-up of milk pressure, though occasionally it may be breached, as in the case of mares that lose milk during late pregnancy (Kainer, 1993).

The alveoli, which are the milk-secreting structures, are lined by a single layer of lactating epithelial cells, surrounding a central

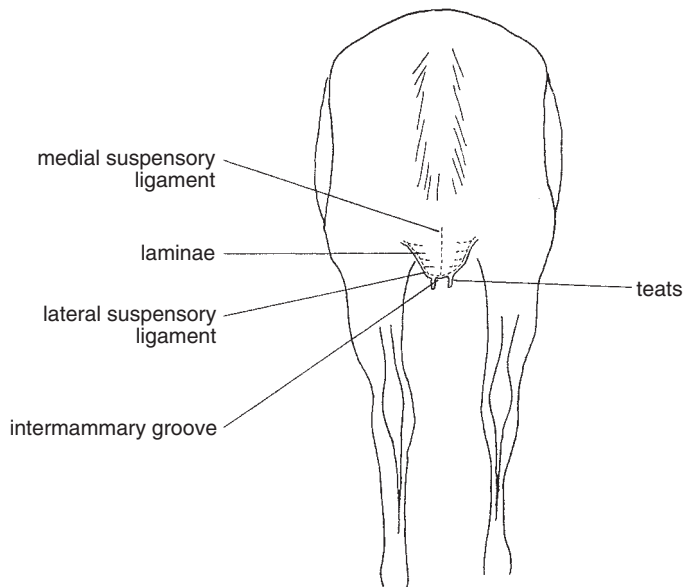


Fig. 9.1. A caudal (rear) view of the mare's udder, illustrating the suspensory apparatus of the mammary gland.

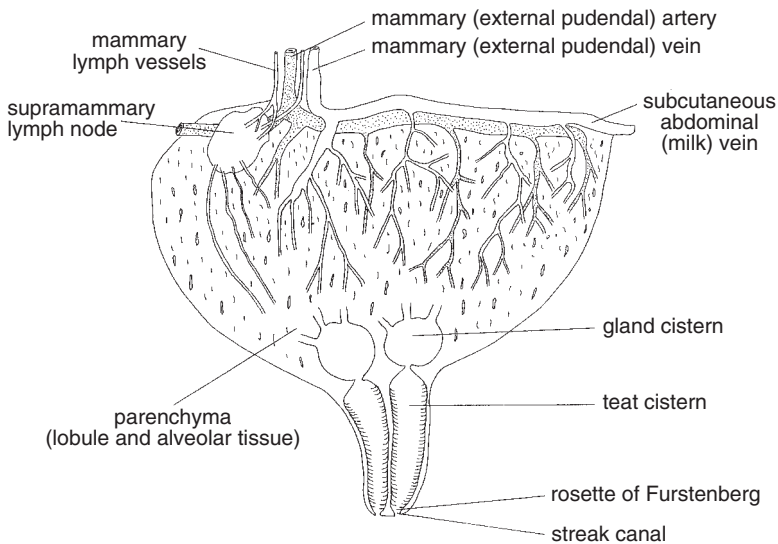


Fig. 9.2. A cross-section through the mammary gland of the mare, illustrating the exit of two quarters via a single teat.

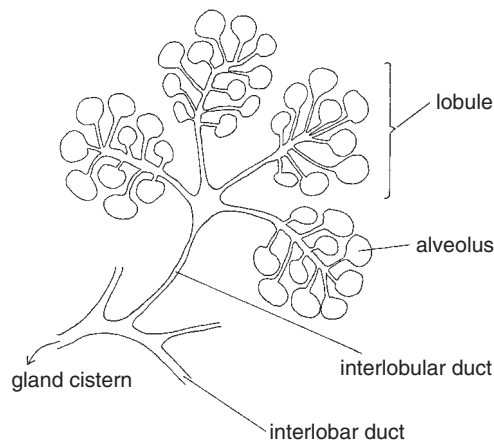


Fig. 9.3. A single lobe of the mammary gland made up of several lobules, which, in turn, are made up of numerous alveoli.

cavity or lumen. This alveolar lumen is continuous with the mammary duct system. Milk is secreted by the lactating cells into the alveolar lumen across the luminal or apical membrane. Surrounding each alveolus is a basket network of myoepithelial

cells. These muscle cells also surround the smaller ducts, and their contraction is activated as part of the milk ejection reflex. Surrounding these myoepithelial cells is a capillary network supplying the alveoli with milk precursors and providing hor-

monal control; there are also a series of lymph vessels. In addition, the alveoli have a nerve supply, which is responsible for the activation of the myoepithelial basket cells and also vasodilatation and constriction of the capillary supply network (Fig. 9.4; Mephram, 1987).

The mammary gland as a whole is supplied with blood via two mammary or external pudendal arteries, one on each side of the midline and entering the caudal end of the gland. Venous return from the mammary gland is via the venous plexus at the base of the gland and then on to the superficial vein of the thoracic wall (the subcutaneous abdominal milk vein) or via the external pudendal vein. Both the external pudendal artery and vein enter and leave the body in the inguinal region. The subcutaneous abdominal vein, which runs along the belly of the mare, can be seen more clearly in lactating mares and is hence sometimes referred to as the milk vein. The mammary gland also has two supramammary lymph nodes, one either side of the midline, at the base of the udder, and connecting the main circulatory lymph system to that of the mammary gland itself (Saar and Getty, 1975).

9.3. Mammogenesis

Mammogenesis, or mammary development, is first evident in the embryo. Glands develop along either side of the midline in the inguinal region. Cells in this region proliferate to form nodules, which develop to form mammary buds, evident from day 50 of gestation. At birth, teats are present, along with a few short branching ducts within the connective tissue associated with each teat.

From birth to puberty, mammary-gland growth is isometric with (at the same rate as) body growth. Most of this prepubertal growth is an increase in fat and connective tissue, rather than duct development. Puberty marks a change, as mammary development becomes allometric, or increases relative to body growth. Beyond puberty, mammary growth increases and decreases with the oestrous cycle. The amount of mammary development within these cycles depends on the length of the dioestrous phase of the oestrous cycle, as elevated progesterone levels are responsible for mammary lobular-alveolar development. In the mare, the duration of dioestrus is relatively long and hence during dioestrus some lobulo-alveolar development does take place.

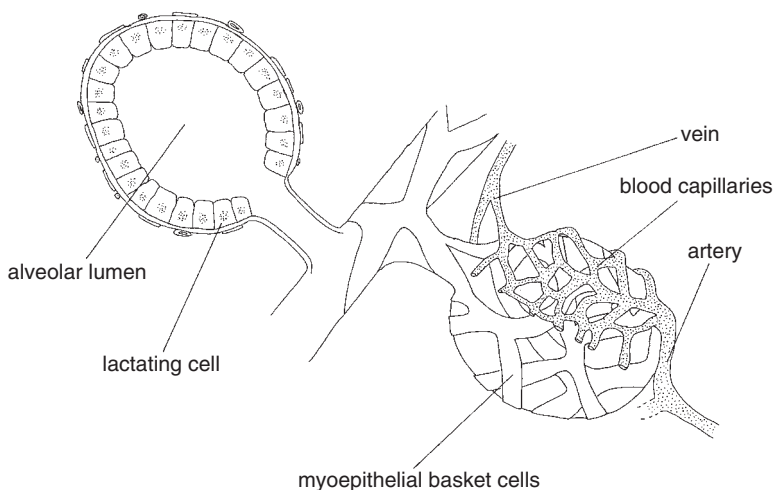


Fig. 9.4. The mammalian alveolus. On the left, a cross-sectional view illustrating the lactating cells surrounding the alveolar lumen, which is continuous with the mammary duct system. On the right, an alveolus illustrating the myoepithelial basket cells and alveolar blood supply.

During pregnancy, elevated progesterone levels again cause significant lobular–alveolar development, especially in the last trimester. In the last 2–4 weeks of pregnancy, lactogenesis (milk production) also predominates (Leadon *et al.*, 1984; Ousey *et al.*, 1984; Mephram, 1987). During lactation, mammo-genesis continues, as cell division increases in line with milk production to satisfy the increasing demands of the foal. Cell division then decreases after the maximum yield has been reached. At the same time, the size of the mammary gland slowly decreases until it returns to its normal non-lactating size postweaning (Fig. 9.5).

9.4. Lactation Curve and Milk Quality

There has been significantly less research conducted on the lactation of the mare compared with other livestock, especially the cow. Except in a very few cultures, mare's milk is of indirect rather than direct commercial importance, its value being assessed via the standard of foal reared, rather than directly by milk yield. As such, it is often not

given the attention it warrants. The following discussion will concentrate on conclusions drawn either directly from experiments using mares or extrapolated from other species studied.

9.4.1. Lactation curve

There is much variation in the lactation curve demonstrated by different individuals, largely due to human interference and early weaning. As a general trend, milk yields in mares tend to increase during the first 2–3 months post-partum. Initial levels in the first 2 weeks are of the order of 4–8 l day⁻¹ for Thoroughbreds and 2–4 l day⁻¹ for ponies (Fig. 9.6). Milk production reflects demand, which in turn reflects the size of the foal; production therefore continues to rise as the foal grows until 2–3 months post-partum, when maximum levels of 10–18 l day⁻¹ in Thoroughbreds and 8–12 l day⁻¹ in ponies are reached (Tyznik, 1972; Oftedal *et al.*, 1983; McCue, 1993). After 3 months, the foal's demand for nourishment from its mother decreases, as it starts to increasingly investi-

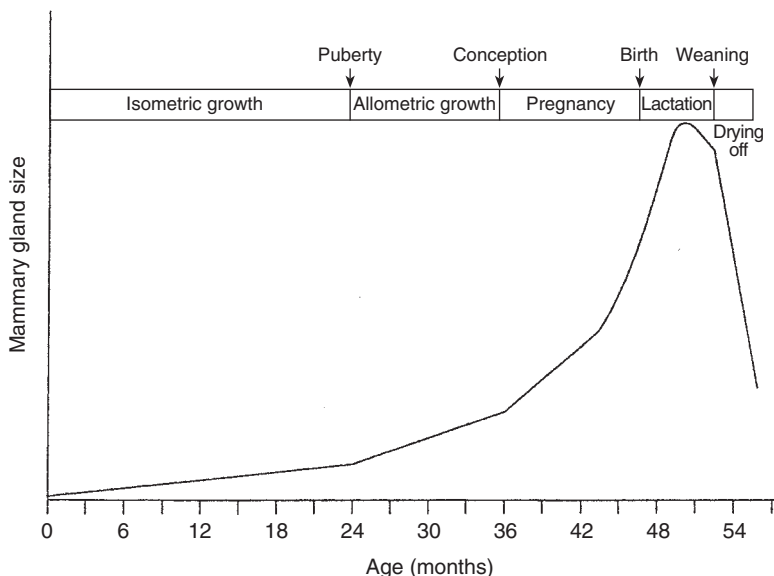


Fig. 9.5. Equine mammary development from birth to 54 weeks, including development during a mare's first pregnancy.

gate grass or hay and its mother's hard feed. As the weaning process progresses towards full weaning, the lactational yield drops off further with decreasing demand (Jacobson and McGillard, 1984; Doreau and Boulet, 1989; Smolders *et al.*, 1990). As will be discussed later, milk quality also declines at this time, further encouraging the foal to seek nourishment elsewhere and hasten the weaning process.

Lactation naturally lasts nearly a full year, the mare drying up completely a few weeks before she is due to deliver the following year. However, in today's managed systems, humans normally dictate the length of lactation by weaning foals at about 6 months, at which time milk yield is less than that immediately post-partum. At this stage the foal is obtaining little of its nourishment from its mother, deriving most from roughage and concentrate feeds. Weaning at 6 months, therefore, has little long-term effect on the foal's development (Fig. 9.6).

The total milk yield of a Thoroughbred or one of the larger riding-type horses is 2000–3000 kg of milk per lactation. As a rough guide, in these larger horses, the natural daily milk yield averaged out over the

whole lactation is 2–3 kg 100 kg⁻¹ body weight. The corresponding equation for ponies is 5 kg 100 kg⁻¹ body weight (Oftedal *et al.*, 1983). The foal normally suckles up to 100 times day⁻¹ during the first week, reducing to 35 times day⁻¹ by week 10. Initial suckling ensures an intake of little but often; however, with age, the frequency of suckling declines and the intake per suckle increases up to 250 g or so at each suckling for larger riding-type horses (Frape, 1998). The number of suckles per day and the amount of milk taken per suckle reduce from peak lactation towards weaning.

9.4.2. Milk quality and composition

The composition of milk reflects the requirement of the young of that particular species and provides the energy and precursors needed for growth throughout lactation. In the case of some mammals, including the foal, milk additionally provides immunoglobulins during the initial stages of lactation (Table 9.1; Ullrey *et al.*, 1966; Peaker *et al.*, 1979; Mephram, 1987; Smolders *et al.*, 1990).

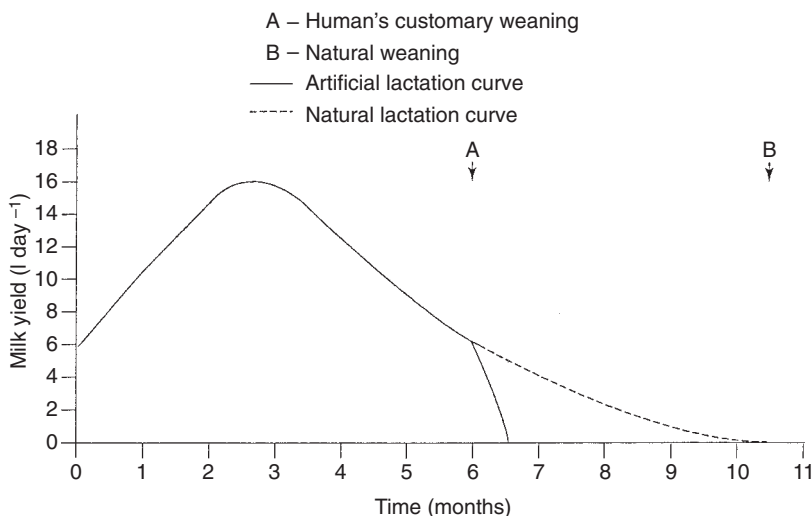


Fig. 9.6. The average lactation curve for a mare, illustrating the natural extent of lactation, along with that customarily imposed by humans.

Table 9.1. Comparative milk compositions of several species, expressed as percentages (from Jennes and Sloane, 1970).

Species	Total solids	Fat	Casein protein	Whey protein	Lactose
Human	12.4	3.8	0.4	0.6	7.0
Cow	12.7	3.7	2.8	0.6	4.8
Goat	13.2	4.5	2.5	0.4	4.1
Sheep	19.3	7.4	4.6	0.9	4.8
Horse	11.2	1.9	1.3	1.2	6.2

9.4.2.1. Colostrum

Colostrum, the first milk, contains a relatively high concentration of proteins, immunoglobulins. Protein concentration in colostrum is of the order of 13.5%, compared with 2.7% in the main lactational milk. The main protein immunoglobulin in mare's colostrum is immunoglobulin G (IgG) (8911 mg dl⁻¹); IgA (957 mg dl⁻¹) and IgM (122 mg dl⁻¹) are of less importance (Kohn *et al.*, 1989; McCue, 1993). This high protein concentration is at the expense of fats, which are present in relatively low concentrations. However, within 12–24 h,

protein levels fall dramatically and fat levels rise. The relative concentrations within milk now stabilize, though both protein and lipid concentrations tend to decline gradually over time. Lactose remains largely unchanged throughout the remainder of lactation (Fig. 9.7; Table 9.2; Ullrey *et al.*, 1966; Forsyth *et al.*, 1975; Gibbs *et al.*, 1982; Smolders *et al.*, 1990). The digestive system of the foal is 'permeable' to the complete protein molecules, such as immunoglobulins, for the first 24 h of life. This 'permeability' is due to enterocytes within the wall of the small intestine, which absorb whole proteins via pinocytosis. After

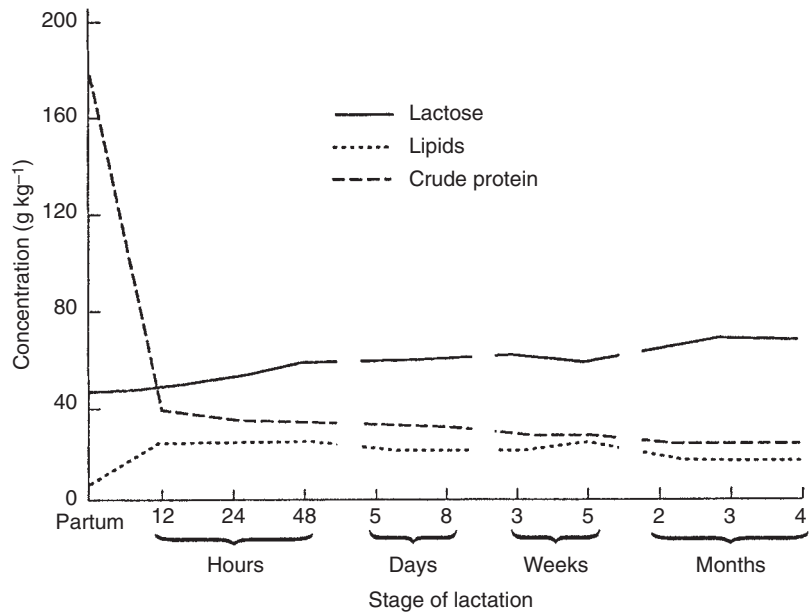


Fig. 9.7. Changes in the concentrations of lactose, lipids and crude protein in milk during lactation (from Ullrey *et al.*, 1966).

Table 9.2. The average composition of the milk during the main part of lactation in the mare (data from Ullrey *et al.*, 1966; Oftedal *et al.*, 1983; Schryver *et al.*, 1986; Saastamoinen *et al.*, 1990; Martin *et al.*, 1991; Frape, 1998).

Component	%
Water (%)	89.0
Protein (g kg ⁻¹)	16–20
Lactose (g kg ⁻¹)	61–69
Fat (g kg ⁻¹)	11–13
Ash (minerals, vitamins, etc.) (g kg ⁻¹)	0.6–3
Calcium (g kg ⁻¹)	0.6–0.8
Phosphorus (g kg ⁻¹)	0.2–0.4
Magnesium (mg kg ⁻¹)	65–90
Potassium (mg kg ⁻¹)	300–500
Sodium (mg kg ⁻¹)	160–200
Copper (μg kg ⁻¹)	200–450
Zinc (μg kg ⁻¹)	800–25,000

24 h, this ability to absorb large protein molecules is irreversibly lost, as the enterocytes are replaced (McCue, 1993). It is essential, therefore, that a newborn foal receives its colostrum well within 24 h of birth, as after this time it cannot take advantage of the immunoglobulins carried by colostrum and they will be broken down by proteolytic enzymes within the intestine into their component amino acids and absorbed as such.

The average composition of milk during the main part of lactation in the mare is given in Table 9.2.

9.4.2.2. Fat

The concentration of fats or lipids in mare's milk is reported to be relatively low when compared with that of other species. However, there is some suggestion that this may be due to sampling error, as the highest concentration of fat is evident in the last milk milked out, which is not easily obtained. Fat is present in milk in the form of globules of saturated fat, cholesterol and unsaturated fats, as free fatty acids, phospholipids and triglycerides. The 8% concentration of triglycerides as a proportion of total fats is much lower than the 79% in cows. These fat globules exist as an emulsion within the milk and contain a high concentration of short-chain fatty acids, fewer than 16 carbons in length.

9.4.2.3. Proteins

Proteins during the main lactation are present in the form of 1.3% caseins and 1.2% whey. Caseins are unique to milk and have several functions. Under the influence of the stomach's acid pH, they form a clot with the enzyme rennin. This clot facilitates the digestion of proteins by the proteolytic enzymes of the digestive system. Caseins also contain essential amino acids and aid in the transport of minerals from the mare to the foal via milk. Caseins associate with calcium (Ca), phosphate and magnesium (Mg) ions to form micelles, thus allowing a higher concentration of these minerals to be transported in milk than would be possible in a simple aqueous solution.

Two types of whey proteins are found in mare's milk and, unlike caseins, do not precipitate in acid pH. The whey proteins are divided into those that are specific to milk and those that can be found in both milk and blood. Those specific to milk can be further subdivided into β-lactoglobulin (28–60% of whey proteins) and α-lactalbumin (26–50% of whey proteins) (Gibbs *et al.*, 1982). α-Lactalbumin is a good source of amino acids and is rich in essential amino acids, such as tryptophan. It is also the B component, which, along with the A component, makes up the two halves of the enzyme lactase synthetase. Lactase synthetase is the

terminal enzyme in the synthesis of lactose, the major sugar component of mare's milk. The second type of whey proteins found in mare's milk constitutes ones also found in blood: serum albumin (2–15% of whey proteins) and serum globulin (11–21% of whey proteins) (Gibbs *et al.*, 1982). Serum albumin is identical to blood serum albumin and is directly transferred unchanged from the blood through the lactating cell to the alveolar lumen. It is, therefore, only found in small concentrations, unless there has been cellular damage or haemorrhage within the mammary tissue. Serum globulin, on the other hand, is the immunological fraction of milk and therefore its concentration is very high in colostrum. Antibodies attach themselves to these globulins and it is via these that the foal attains its passive immunity.

9.4.2.4. Lactose

Lactose is the energy component of mare's milk (6.1%). Unique to mammals, each lactose molecule consists of a molecule of galactose and one of glucose. In the intestine of the foal, lactose is split into its two component parts, galactose then being easily converted into glucose. Lactose is therefore, in essence, two molecules of glucose. The question then arises as to why lactose, not glucose, is present in milk, especially as there is an energy cost in converting glucose to lactose and vice versa. The answer lies in the effect of glucose on the osmotic pressure of milk relative to blood. The osmotic pressure of the two must be the same, and the component of milk that has the largest effect on osmotic pressure is the small molecule of lactose. However, if glucose were present, it would have an even greater effect on the difference in osmotic pressure. Additionally, one molecule of lactose gives rise to two molecules of glucose; that is, one molecule of lactose has twice the calorific value per molecule compared with glucose, and hence also per unit of osmotic pressure. It has also been suggested that lactose provides a more beneficial medium for intestinal activity, regulates bacterial flora and stabilizes pH, so aiding the absorption of minerals (Mephram, 1987; Smolders *et al.*, 1990; McCue, 1993).

9.4.2.5. Minerals

Mineral concentrations also vary with the stage of lactation. Potassium (K) and sodium (Na) concentrations in colostrum tend to be high, at 1200 mg kg⁻¹ and 500 mg kg⁻¹, respectively, dropping to 600 mg kg⁻¹ and 200 mg kg⁻¹ within 5 weeks. Ca tends to be slightly raised in colostrum, but then falls away slightly within hours only to rise again to a peak of 1200 mg kg⁻¹ at 3 weeks post-partum. Mg levels are also elevated, at 500 mg kg⁻¹, in colostrum but fall off rapidly in the first 12 h and then continue to decline slowly throughout the rest of lactation. Phosphorus (P) remains relatively steady, at 400–500 mg kg⁻¹, for the first 8 weeks of lactation and then concentration declines (Figs 9.8 and 9.9; Ullrey *et al.*, 1966; Oftedal *et al.*, 1983; Schryver *et al.*, 1986; Saastamoinen *et al.*, 1990; Martin *et al.*, 1991).

It is evident that there is a trend for the concentration of all the nutrient components of milk to decline as lactation proceeds. This is nature's way of encouraging the foal to obtain its nourishment elsewhere (Smolders *et al.*, 1990).

9.5. Milk Synthesis

Milk is synthesized in the epithelial or lactating cells lining each alveolus. The precursors of and components for milk are obtained from the blood system supplying the udder. These components cross the basal membrane into the lactating cells. There is little information on how they pass across this membrane, but, as the molecules are small, it seems likely that it is by diffusion. The protein, fat and lactose components of milk are then built up within the lactating cells and pass across the cell membrane to the lumen of the alveolus (Mephram, 1987). Each of the major components of milk is discussed below.

9.5.1. Proteins

Proteins are built up from amino acids within the lactating cells. The total amount of nitrogen that crosses the basal membrane

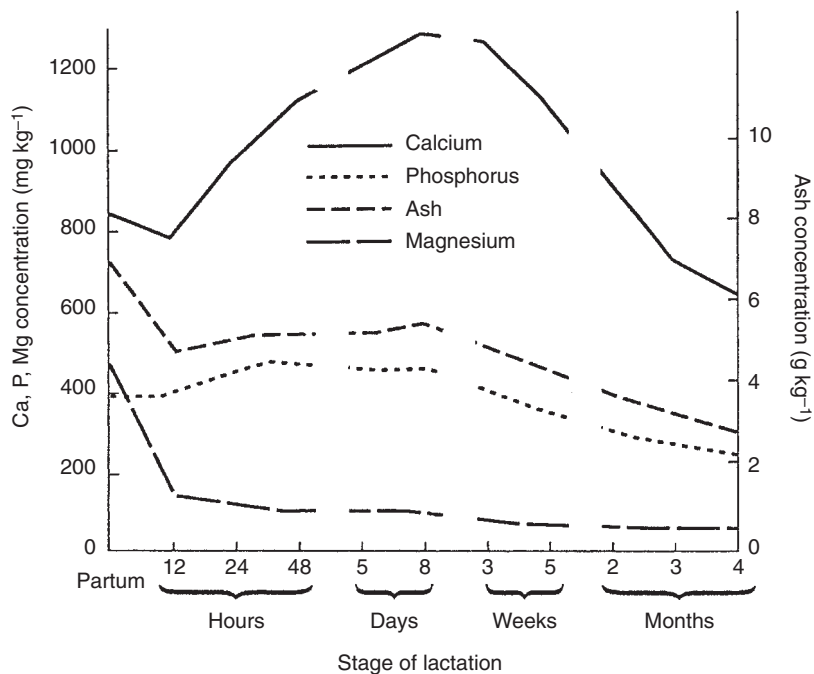


Fig. 9.8. Changes in the concentrations of calcium, phosphorus, magnesium and ash in mare's milk during lactation (from Ullrey *et al.*, 1966).

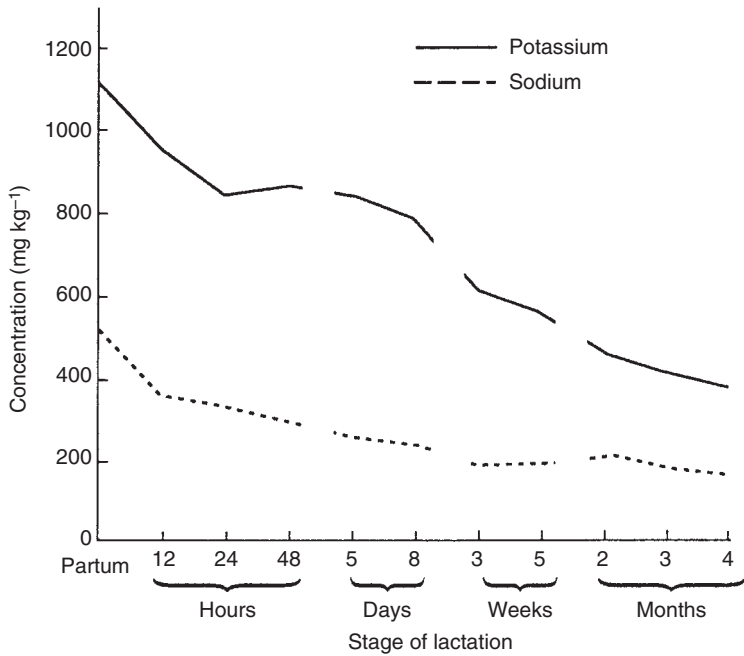


Fig. 9.9. Changes in the concentrations of potassium and sodium in mare's milk during lactation (from Ullrey *et al.*, 1966).

is equal to that within milk; however, there is a change in amino acids. Non-essential amino acids are synthesized within the cell and are built up into proteins along the mRNA within the ribosomes of the rough endoplasmic reticulum (RER). These proteins are then secreted into milk. Essential amino acids are passed unchanged across the basal membrane of the lactating cell and are incorporated into proteins, along with the non-essential amino acids synthesized within the cell.

9.5.2. Lactose

Blood glucose is the primary precursor of lactose. However, glycerol, acetate and amino acids are also thought to contribute. The amount of glucose absorbed by the gland is much more than is needed solely for conversion to lactose. The difference is used as energy for general cell metabolism. The conversion of glucose to lactose involves five enzymes, the fifth enzyme being lactose synthetase, which is made up of two components, A and B. As discussed previously, component B is the major milk protein, α -lactalbumin. The biochemical pathways involved in the conversion of glucose to lactose are summarized in Fig. 9.10.

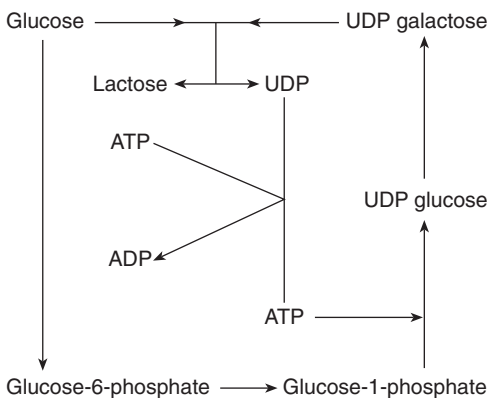


Fig. 9.10. A summary of the conversion of glucose to lactose within the lactating cell. UDP, uridine diphosphate; ATP, adenosine triphosphate; ADP, adenosine diphosphate.

9.5.3. Fat

The fat globules within milk are made up of esterified glycerol and free fatty acids, which aggregate to form a fat-droplet emulsion within milk. There is much variation in the length of free fatty acids making up the fat globules in the milk from females of varying species. The horse tends to have a higher concentration of short-chain fatty acids (fewer than 16 carbon (C) atoms in length).

Fatty acids are derived mainly from three sources: glucose; triglycerides; and free fatty acids. Glucose C is a significant precursor of free fatty acids in the non-ruminant – for example, the horse. Glucose is absorbed across the basal membrane and converted to acetyl coenzyme A (CoA) and on to malonyl CoA within the cytosol of the cell. Malonyl CoA is then built up, using a multienzyme complex, to free fatty acids, which tend to be short-chain (<16C). Blood triglycerides provide an alternative source of free fatty acids for the lactating cell; these are broken down into glycerol plus free fatty acids within the cell. The free fatty acids obtained from triglycerides are longer-chain fatty acids, typically 16–18 C in length. Triglycerides, therefore, are not a very important source of fatty acids in the mare. The triglycerides are either broken down into amino acids and glycerol in the blood, similarly to the way that proteins are broken down into amino acids, and then absorbed into the cell, or they are absorbed directly.

Glycerol that combines with the free fatty acids is derived again by three different methods: from the breakdown of triglycerides within the cell; by the absorption of free glycerol in the blood; or, finally, from the breakdown of glucose within the cell.

The free fatty acids and glycerol within the cell combine by esterification within the endoplasmic reticulum. These molecules then aggregate together to form the fat droplets within milk.

9.6. Milk Secretion

All the components of milk produced by the lactating cells have to pass across the apical membrane of the lactating cell into the alveolar lumen (Fig. 9.11). The different components of milk pass by different mechanisms.

9.6.1. Fat

The fat-droplet size increases as the free fatty acids and glycerol continue to combine by esterification and the resulting molecules aggregate into increasingly larger droplets as they migrate towards the apical membrane. In the vicinity of this membrane, strong London–van der Waals forces attract these fat droplets and envelop them in the membrane, forming a bulge in the apical membrane surrounding the droplet (Fig. 9.12).

The droplet and surrounding plasmalemma move away from the apical membrane into the lumen, forming a narrow bridge. This bulge then pinches off as the bridge gets narrower and releases the fat droplet plus surrounding plasmalemma into

the alveolar lumen. The process is termed pinocytosis. Occasionally, part of the cell cytoplasm, sometimes including cell organelles, is enclosed in the bulge of the apical membrane along with the fat droplets, and then gets secreted into the alveolar lumen along with the milk fat. The formation of these structures, termed signets, occurs more in the lower order of mammals, but they are occasionally evident in mare's milk (Mephram, 1987).

9.6.2. Protein

Proteins are built up from their constituent amino acids along the RER within the cell and then pass on to the Golgi apparatus. They accumulate as granules of proteins within the Golgi; this Golgi apparatus then migrates towards the apical membrane. The membrane of the Golgi apparatus fuses with the apical membrane and this releases the proteins into the alveolar lumen by reverse pinocytosis (Fig. 9.13). By this reverse pinocytosis, plasmalemma lost during the secretion of milk fat is replaced during the secretion of milk proteins.

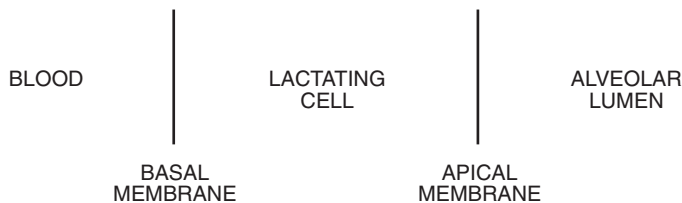


Fig. 9.11. The route of passage for all milk components from the mare's blood supply on the left through the lactating cell to the lumen of the alveolus.

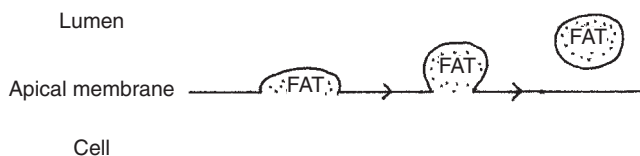


Fig. 9.12. The secretion of milk fat from the lactating cell (below) into the alveolar lumen (above) by pinocytosis.

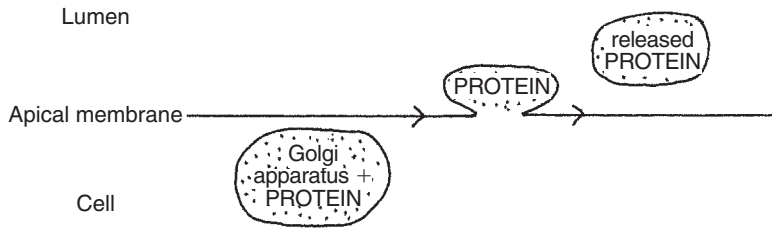


Fig. 9.13. The secretion of milk protein from the lactating cell (below) into the alveolar lumen (above) by reverse pinocytosis.

9.6.3. Lactose

The secretion of molecules of lactose, unlike that of milk fat and protein, is not visible using electron microscopy, and so the method of secretion is less clear. As discussed previously, one of the mare's milk proteins is α -lactalbumin, and this protein is the B component of the fifth and last enzyme involved in lactose synthesis. It therefore seems likely that lactose secretion is closely linked to that of milk protein. The A protein of the enzyme lactase synthetase is known to be closely

associated with the membrane of the Golgi apparatus. The B component (α -lactalbumin) is synthesized, as are all other milk proteins, on the RER and then passed on to the Golgi apparatus. While the B component is in the Golgi apparatus, it becomes associated with the A component already there, and together they form active lactase synthetase. This enzyme catalyses the conversion of uridine diphosphate galactose and glucose to lactose. The lactose is then presumed to be secreted along with the milk proteins by reverse pinocytosis (Figs 9.13 and 9.14).

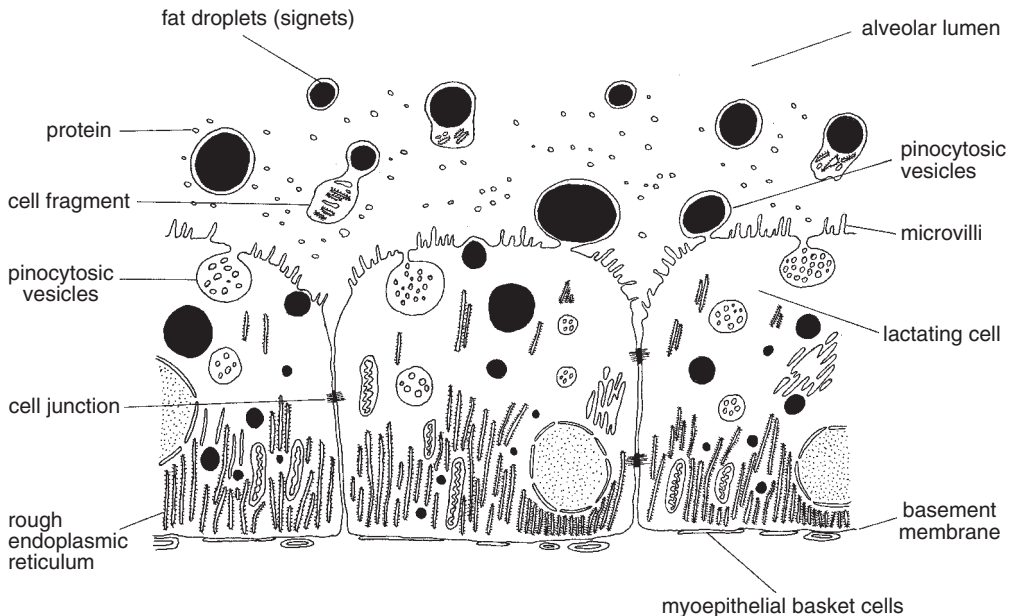


Fig. 9.14. Diagrammatic representation of a mammary secretory cell illustrating the build-up of protein and fat for release into the alveolus.

9.6.4. Minerals

Milk has a relatively high concentration of K when compared with Na, and this is similar to the relative concentrations within the cytoplasm of the cell. Na and K in milk are derived from the intercellular fluid. The components within the lactating cell are derived from the blood system but the K:Na ratio in blood is the reverse of that within the cell and milk. Therefore, there must be an active transfer system across the basal membrane. This is via a Na pump, which pumps Na away from the cell cytoplasm and into the blood system and pumps K the opposite way towards the cell. This maintains the high K:Na ratio within the cell. As the K:Na ratio in the milk is the same as that in the cell, then Na and K are presumed to pass across the apical membrane to the alveolar lumen by simple diffusion (Mephram, 1987).

The concentrations of Ca, Mg and P are higher in milk than in the cell cytoplasm. Therefore, their passage must be via an active transport system. The exact mechanism is unclear but all three ions are known to be closely associated with the milk protein casein. It is therefore assumed that this association occurs within the Golgi, where the casein proteins are synthesized. These ions are then passed into the milk, along with proteins, via reverse pinocytosis (Mephram, 1987).

Iron is also secreted in association with proteins by reverse pinocytosis, as it is specifically bound to a minor milk protein, lactoferrin (Mephram, 1987).

9.6.5. Water

Water passes to the alveolar lumen from the cell cytoplasm by osmotic pressure. Fat and

protein molecules in milk are in the form of large droplets, and so their effect on osmotic pressure is minimal. However, lactose and free ions are much smaller and it is these that affect osmotic pressure and hence drive water diffusion from the cell into milk (Mephram, 1987).

9.6.6. Immunoglobulins

Colostrum, as discussed previously, has a high concentration of proteins; these proteins are immunoglobulins and their associated antibodies. These immunoglobulins are combined into large corpuscles, termed bodies of Donné. The mechanism by which these are secreted is unclear. It is possible that engorgement of the lactating cell in late pregnancy results in the breakage of some of the junctions within the cell membranes, especially the basal cell membranes of the lactating cells. This allows the serum proteins to pass into milk unchanged. There is also evidence suggesting an active transport system for these serum proteins, but the exact mechanism is as yet unclear.

9.7. Conclusion

Our present knowledge specifically regarding equine lactation is still limited, however, by extrapolation from other species, a reasonable understanding can be achieved. Caution must be practised, however, in making definitive statements until these assumptions and extrapolations have been confirmed or refuted when more detailed research, specifically on equine lactation, is conducted.

10

Control of Lactation in the Mare

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10.1. Introduction

The control of lactation in most mammals, including the mare, is via both nervous and hormonal pathways. Unfortunately, information specific to the mare is very limited, though it is assumed that the control of lactation is very similar to that evident in other mammals. The information discussed in this chapter is gleaned from the limited experiments carried out on horses and extrapolation from other mammals where appropriate.

Lactation may be divided into three stages as far as its control is concerned: lactogenesis; galactopoiesis; and milk ejection.

10.2. Lactogenesis

Lactogenesis refers to the initial milk secretion that occurs in late pregnancy prior to parturition, resulting in a build-up of colostrum within the mammary glands. In

the mare, lactogenesis is evident well before parturition occurs, as demonstrated by the presence of lactose, proteins and fat within the mammary secretions (Peaker *et al.*, 1979).

Control of lactogenesis is hormonal. High progesterone concentrations characteristic of pregnancy drive lobular-alveolar development, but inhibit milk secretion. Hence, with the decline of progesterone in late pregnancy inhibition of milk production is removed (Mephram, 1987). In addition, elevated prolactin, growth hormone and cortisol actively drive milk production. It appears that in the mare prolactin may play a significant role, as concentrations are observed to increase in the last 2 weeks of pregnancy (Neuschaefer *et al.*, 1991). Lactogenesis therefore increases in late pregnancy and reaches a maximum immediately prior to parturition (Forsyth *et al.*, 1975; Peaker *et al.*, 1979; Worthy *et al.*, 1986).

In other mammals – for example ruminants and humans – a placental lactogen has been identified and found to have an

additional effect on lactogenesis. No such placental lactogen has been identified in equines (McCue, 1993).

10.3. Galactopoiesis

Galactopoiesis is the term given to the maintenance of milk production. Again, little information specific to the horse is available. However, it is assumed that control is similar to that in the sheep and the cow, where it is under the control of prolactin, growth hormone and cortisol, increasing concentrations of which act to drive galactopoiesis (Neuschaefer *et al.*, 1991). Galactopoiesis is driven by, and mimics, the foal's demand for milk, and this, in turn, dictates and governs the shape of the lactation curve and the quantity of milk produced.

10.4. Milk Ejection

Milk ejection, also termed milk let-down, differs from the other stages of lactation in that its control is both neural and hormonal. A nervous reflex acts as the stimulus or afferent pathway and hormones form the efferent path. Nerve receptors within the teats are stimulated by the action of suckling, and the nervous afferent pathway is activated, resulting first in a localized effect, causing localized myometrial cell contraction. Second, this afferent nervous pathway acts via the central nervous system (CNS) to stimulate the paraventricular nucleus within the mare's hypothalamus. The hypothalamus then activates the posterior pituitary, which in response produces the hormone

oxytocin. The efferent pathway of the milk-ejection reflex is formed by the oxytocin, which passes into the systemic blood system and hence to the mammary gland. The effectors that react to oxytocin are the myoepithelial basket cells surrounding each alveolus and the small ducts, causing them to contract further and forcing milk out of the alveoli, along the ducts, to the gland cistern and on to the teat cistern, ready to be removed by the suckling action of the foal (Ellendorff and Schams, 1988; Nett, 1993b). Hence, at suckling, the milk initially available is that within the gland and teat cisterns, which is removed by the negative pressure exerted by the suckling action of the foal. This is then closely followed by the milk-ejection reflex, which replenishes the milk within the gland and teat cisterns, making more available to the foal.

In addition to the above control mechanisms, the CNS has an overriding effect. For example, stress, especially as a result of fear or shock, reduces the effectiveness of the milk-ejection reflex by increasing the levels of circulating adrenalin. Adrenalin causes vasoconstriction, so reducing the amount of oxytocin reaching the alveoli and hence the effectiveness of the reflex (Fig. 10.1).

10.5. Conclusion

It is evident that there is much left to be learnt about the control of lactation in the mare. The lack of direct commercial value has led to little research into equine milk production and, as such, many extrapolations and assumptions from the cow have been applied to the horse.

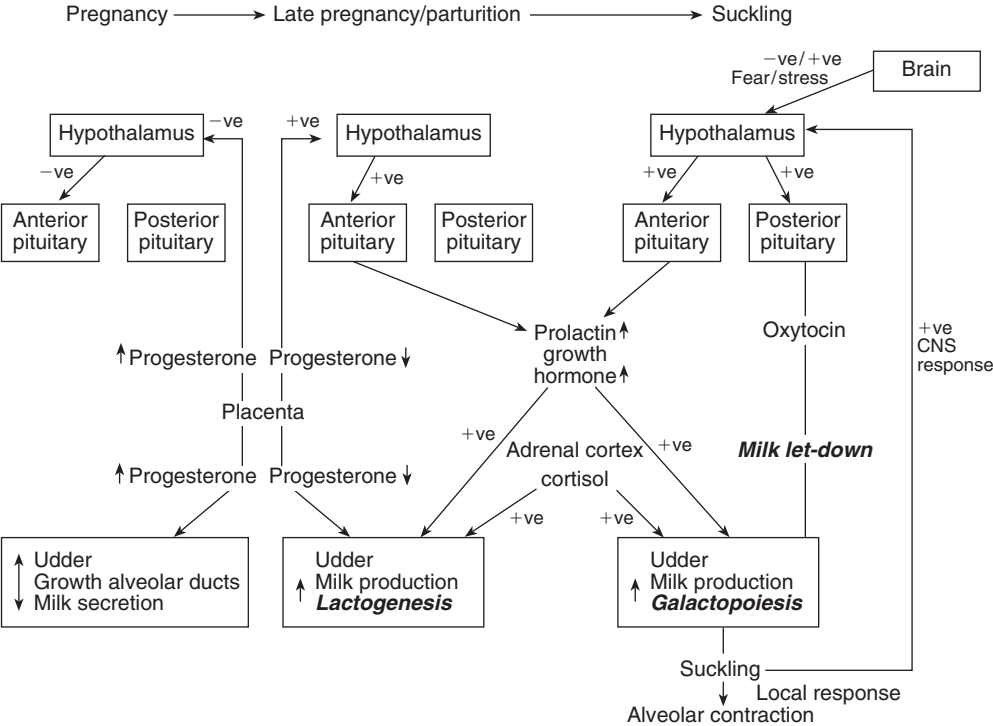


Fig. 10.1. Schematic representation of the control of lactation in the mare. CNS, central nervous system.

11

Selection of the Mare and Stallion for Breeding

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11.1. Introduction

The choice of both mare and stallion for breeding can be a very complicated and time-consuming process. Often not enough importance is placed upon this selection, the result of which is an oversupply of mediocre or poor stock and unnecessary difficulties with mares at covering, during pregnancy, at foaling and during lactation.

One of the most obvious selection criteria is that of performance or athletic ability. If you are looking to produce a quality show horse, you will obviously be selecting for conformation and show success on both the mare's and stallion's side and in their breeding. If you are looking to produce a racehorse or competition horse, you will be interested primarily in the proven performance of the dam and sire and select accordingly. Finally, if you are looking for a leisure horse, you will be interested primarily in temperament and possibly hardiness.

It is beyond the scope of this book to discuss in detail the wide range of performance criteria that a breeder may look for in order to achieve that specific, ideal horse. These performance criteria vary considerably with the individual breeder and the type of horse required. Many books and articles have been written on the subject, but ultimately it is a personal decision.

Regardless of the performance criteria used for selection, stock should also be selected on reproductive competence. However, all too often, such criteria are not considered, with potentially serious consequences for the individual breeder and the equine breed as a whole. Regardless of the type of horse you intend to breed, reproductive competence and the ability to produce healthy offspring with minimal danger to the life and well-being of the dam should also be of prime importance. Today's horse, unlike other farm livestock, has been selected primarily for performance ability, often at the expense of reproductive competence. As a result, there are many potential reproductive problems that the breeder should be aware of in selecting both the mare and the stallion.

The following sections will concentrate solely upon the criteria and techniques that can be used in the selection for reproductive competence and will assume that the selection criteria for performance and conformation have already been met. All the possible techniques will be included in the following sections, many of which are costly in time and money. The extent to which these techniques are used depends on personal choice and the value of the breeding stock concerned and the potential offspring. Further information specific to infertility and hence an expansion of some of the issues raised here are included in Chapter 19.

11.2. The Mare

The selection criteria for reproductive competence in the mare can be listed as follows:

- History
- Temperament
- Age
- General conformation and condition
- External conformation of the reproductive tract
- Internal conformation of the reproductive tract
- Infections
- Blood sampling

11.2.1. History

The history of the mare includes both her specific breeding history and her general history (Shideler, 1993a). Many mares these days, especially those of any value, come with full historical documentation. Such records are invaluable in assessing her ability to produce a foal, as well as easing management. If there are no records available, contact should be made with her previous owners to find out as much information as possible.

11.2.1.1. Reproductive history

Details of her past breeding performance should ideally include answers to the following questions: Does she show regular

oestrous cycles? What is the length of her normal oestrous cycle? When does her breeding season normally start? How long does her season normally last? Does she show oestrus well and are there any characteristic signs? How long is her typical oestrus? Does she demonstrate oestrus better under certain circumstances? etc.

This information will indicate, among other things, whether or not she will be easy to detect in oestrus and cover. Mares that do not demonstrate oestrus well tend to be hard to cover. This leads to frustration, due to missed oestruses and wasted journeys to the stallion, all increasing costs. In addition, mares that are habitually hard, or even dangerous, to cover may be refused by some studs, reducing the pool of potential stallions or necessitating the use of artificial insemination (AI).

Other questions asked should include whether she has bred before. If so, has she had problems holding to the stallion? This will necessitate return journeys to the stud or a prolonged period of time away. Has she had problems during any of her pregnancies? Has she ever reabsorbed or aborted? Does she or her family throw twins? If a mare has a history of habitual reabsorption or abortion or the need for holding injections (artificial progesterone supplementation) post service, then she is really not a good candidate for a brood mare. Such problems may indicate an inherent inability to carry a pregnancy to term, due to hormonal imbalance, uterine incompetence and/or genetic abnormality.

Repeated incidences of twins are a problem, in that the likelihood of one or both of the fetuses aborting is very high. If only one of the twins aborts, the remaining one is often born weaker and smaller than would normally be expected. Spontaneous abortion of twins is not due to an inherent inability to carry a foal to term and such mares are perfectly capable of producing a foal providing a single pregnancy is conceived. They may, however, need to be checked routinely for twin pregnancies and managed appropriately to eliminate one twin at an early stage or to induce abortion, with the mare being returned to the stallion (Ginther, 1982; see Chapter 14).

It is important to find out whether there may have been any incidences of dystocia. It is rare for abnormal fetal positions to occur repeatedly, but past difficulties may have caused internal lacerations or damage. It is particularly important to know if the cervix has been affected. Limited damage to the uterus, vagina and vulva will repair quite effectively, though it may leave areas of weakness or adhesions. Damage to the cervix may cause cervical incompetence, allowing bacteria to enter the higher reproductive tract and thus causing infection. Conversely, adhesions may hinder the passage of sperm at covering and make the cervix less able to dilate during parturition, necessitating a Caesarean delivery.

Previous post foaling problems may also be indicated, including rejection or even habitual attacking of foals. This could be a sign of a general temperament fault, or may not show up in subsequent pregnancies, particularly if such behaviour was associated with a first foaling. Records may show that the mare is not a good mother, producing ill-thrifty foals. It is therefore helpful to look at foal birth weight and subsequent development and growth rates. Such effects may be due to poor milk yield, which in turn may reflect faults in nutritional management rather than a specific mare problem, but, if she has consistently produced foals that did not do well, it may be worth investigating further.

Records should also show any incidences of infections and the treatments given. Minor infections may show no long-term effects. However, infections of the reproductive tract are largely responsible for the relatively high infertility rates in horses and are the single major cause of fertilization failure and abortion in mares (McKinnon and Voss, 1993). Previous mastitic infections, though rare, should be considered and such mares examined to make sure that the udder has not suffered permanent damage.

Detailed mare records not only prove invaluable as an aid to selection; they also provide useful information for the stud to which you intend to send your mare. This is especially important if she is to stay at stud for a prolonged period of time and oestrus detection is to be carried out by the stud.

11.2.1.2. *General history*

In addition to specific breeding records, information on the mare's general history is also very useful. These should indicate her vaccination and worming status, as well as accidents, especially if these involved injury to the pelvic area, abdominal muscles, internal injuries or damage to limbs or muscles. These may be exacerbated by pregnancy, especially in the later stages, and may predispose the mare to problems or prevent natural parturition.

Ongoing or past conditions should also be noted. Mares with a history of respiratory or circulatory disorders, chronic obstructive pulmonary disease, exercise-induced pulmonary haemorrhage, umbilical hernias or vaginal prolapse would possibly not stand up to the strain of pregnancy. Such conditions may also be exacerbated by pregnancy itself (Turner and McIlwraith, 1982; Shideler, 1993 a,b). Severe laminitis, navicular disease, tendonitis, etc. may also have a bearing on the ability of a mare to carry a pregnancy to term (Woods, 1989). Mares with disorders that are potentially heritable may be able to carry a pregnancy to term, but it is debatable whether such mares should be bred and hence perpetuate a problem within the population.

11.2.2. **Temperament**

The temperament of the mare is obviously important for ease of management, especially at birth and with a young foal, when many mares tend to be antisocial. In addition, temperament is an inherited characteristic and hence that of the parents has a long-term effect on the temperament and character of the foal. The mare in particular has a significant effect on foal temperament, due to their prolonged proximity during the foal's early life until weaning.

A mare with a quiet and gentle disposition is much easier to handle and manage. Such mares tend to show oestrus more readily and are therefore easier to cover. Anxious and highly-strung mares with poor temperaments may show signs of aggression

towards a teaser or stallion even though she is in oestrus; stress may often cause masking of oestrous signs in such mares. Such aggressive mares are obviously more difficult and dangerous to cover. Many need to be twitched and/or hobbled, which further increases stress and is not conducive to optimum fertilization rates or easy management. Such mares can, of course, be covered using AI, but the problem of detecting oestrus remains. Highly-strung and nervous mares may also suffer from higher abortion rates. Finally, a masculinized temperament may be indicative of hormonal abnormalities – for example, granulosa-cell tumours (Hinrichs and Hunt, 1990).

11.2.3. **Age**

The age of a mare at her first breeding does influence the ease of her pregnancy and may have potential long-term effects, especially in mares under 5 or over 12 years of age (Day, 1939). As mares reach puberty between 18 and 24 months of age, it is theoretically possible to breed a mare at 18 months to produce a foal at 29–30 months of age. However, evidence, especially from research into sheep, demonstrates that embryo mortality rates are significantly higher in maiden pubertal animals, and return rates in young maiden mares do tend to be higher than in older multiparous mares.

Young mares may not only be hard to get in foal but they may suffer detrimental long-term effects if nutrition is inadequate. A horse does not attain its mature body size until, on average, 5 years of age, so, for a mare under 5 years, there are the additional demands of growth on top of those of pregnancy and maintenance. This should be reflected in her nutritional intake. To breed a mare early in life is a decision not to be taken lightly. She must be well grown for her age and in a good, but not overfat, physical condition. There must also be the wherewithal to feed her additional good-quality food, especially in late pregnancy, and to provide good accommodation for her in order to minimize her body's maintenance requirement.

At the other end of the spectrum, maiden mares over the age of 12 may also find it difficult to carry a pregnancy and bring up a foal. There is evidence to suggest that embryo survival rates, as well as fertilization rates, are reduced in old mares (Ball, 1988, 1993a; Ball and Brinsko, 1992). This is due to an increase in the incidence of embryonic defects, a reduction in ova viability and an increase in age-related endometritis (see Chapter 19; Ball, 1988, 1993a; Ricketts and Alonso, 1991). In addition, the mere fact that she has been alive longer also means that she has had a greater chance of exposure to reproductive-tract infections, which, if severe, may have permanently affected her ability to conceive. Older mares may also have spent the majority of their lives as barren mares without the attentions of the stallion; they tend, therefore, to reject or be antagonistic towards a stallion's approaches.

Older maiden mares may also have problems associated with their previous life. Many such mares are ex-performance horses and, as such, they have been trained and kept in top athletic condition, which often disrupts reproductive activity. Prime athletic condition is associated with a variety of reproductive malfunctions – for example, delayed oestrus, prolonged dioestrus, complete reproductive failure, etc. These mares need a readjustment period before they are physically and psychologically capable of conceiving, bearing and rearing a foal. A prolonged period of rest, at least 6 months, allows the mare's system to settle down into a non-athletic state; some mares may take as long as 18 months to adjust to their new way of life. If a mare has been treated with drugs, such as corticosteroids and anabolic steroids, illegally or legally, her system will require time to eliminate them, during which time reproductive function may continue to be disrupted (Shoemaker *et al.*, 1989). A performance horse, in top athletic condition, develops musculature, especially in the abdominal and pelvic region, not necessarily conducive to an easy pregnancy and parturition. Time should therefore be allowed for muscle tone to relax.

The ideal age to breed a maiden mare is at 5–6 years of age, at which time she will have reached her mature size. She will also not be old enough to have become set in her ways and will be less likely to have developed aggressive tendencies towards the stallion and to have contracted uterine infections. However, breeding at this age will not suit all systems. Most mares will not have proved their worth so the decision to breed may not yet have been made. One increasingly popular way of overcoming this is the use of embryo transfer, discussed in detail in Chapter 21, which allows performance mares to breed but not at the expense of their performance careers.

Once a mare has borne one foal, she is more able to cope with the demands of subsequent pregnancies and, as such, is much more likely to breed successfully as an older mare well into her teens (Allen, 1992). If a mare has been a brood mare all her life, then age is less important. She may naturally be barren during the occasional year. This has been suggested as nature's way of allowing recovery and regeneration. More care and attention are needed with older age, and it is not normally advisable to breed a mare over 20 years old, though this largely depends on the breed, type and condition of the individual mare. Mares of the native type tend to breed more successfully into later life than the hot-blood/warm-blood-type animals, though of course there are exceptions to every rule.

11.2.4. General conformation and condition

A mare's general conformation is of importance, not only to ensure that her offspring are well conformed, but also to ease her pregnancy. She needs to have a strong back and legs to enable her to carry the considerable extra weight of the fetus during late pregnancy. She should have correct pelvic conformation, with a pelvic opening adequate for a safe delivery. Ideally, she should also possess good heart and lung room across the chest and have plenty of abdominal space. In general, rather fine, tucked-up

mares tend to be poorer breeders or have more problems during pregnancy.

A mare's general body condition is considered to have implications for reproductive activity, though evidence is conflicting. The body condition of horses can be classified on a scale of 0–5, 0 being emaciated and 5 obese. The ideal condition score for a mare at mating is 3. Such mares have a good covering of flesh; the ribs may be felt with some pressure, as can the vertebrae of the backbone. It is widely believed that mares with condition score 3 have the highest fertilization rate and subsequent reproductive success (Fig. 11.1;

Van Niekerk and Van Heerden, 1972; Kubiak *et al.*, 1987; Morris *et al.*, 1987).

Thin mares may show prolonged anoestrus or very long/delayed oestrous cycles, along with suspension of all reproductive activity in cases of emaciation. At the other end of the spectrum, over-fat mares (Fig. 11.2) may suffer reproductive failure, due to excess fat deposition on the reproductive tract limiting their ability to move and expand with a developing pregnancy. Fat may also be deposited on the ovaries and around the Fallopian tubes, interfering with the process of ovulation and ova transfer.



Fig. 11.1. Mare in condition score 3, ideal condition for covering.



Fig. 11.2. Mare in over-fat condition, condition score 5.

It has been demonstrated that, in barren mares, an increasing plane of nutrition 5–6 weeks prior to mating, with an accompanying gradual improvement in body condition up to a score of 3, results in the best ovulation rates and reproductive success (Van Niekerk and Van Heerden, 1972). This process is termed flushing and is a common practice in sheep and cattle management. It is discussed further in Chapter 12. Finally, work done by Swinker *et al.*, (1993) indicates that body condition can also affect the mare's response to hormonal treatment used in the manipulation of reproduction.

11.2.5. External examination of the reproductive tract

Poor external conformation of the mare's reproductive tract can have severe implications for reproductive performance (see Chapter 1).

The perineal area forms the outer vulval seal of the tract (see Chapter 1) and so should be examined. First, the presence of lacerations, damage, puckers, scars, etc. should be noted, as they may be indicative of more extensive internal damage. Second, the general conformation of the perineal area

should be assessed. If the vulval seal is incompetent, this allows bacteria and air-borne pathogens to enter the vagina and challenge the upper reproductive system (see Chapter 1; Figs 1.4, 1.6 and 1.7). This predisposes the mare to pneumovagina and urovagina. Both of these are common causes of reproductive-tract infection and hence infertility. Such conditions are prevalent in Thoroughbred mares, mares in poor condition and mares with reduced vulval tone due to injury, damage, age, oestrus or foaling (Pascoe, 1979a).

As discussed in Chapter 1, the height of the pelvic floor also has a bearing on bacterial contamination and so should also be considered when selecting mares. It may be assessed by inserting a sterile probe into the vagina and resting it on the pelvic floor (Fig. 11.3). In a normal conformation, at least 80% of the vulva should lie below the pelvic floor in order for the vaginal seal to be fully competent. In a mare with a low pelvic floor, the risk of contamination of the reproductive tract can be reduced if perineal conformation is correct and hence the vulval seal is competent.

The external perineal conformation and hence vulval seal competence can be assessed by eye. A simple ruler and pro-

Fig. 11.3. Diagram to illustrate the assessment of vaginal and vulval competence and hence the likelihood of pneumovagina.

tractor can be used to allocate a Caslick index (see Chapter 1) and so indicate the likelihood of infection and the need for a Caslick vulvoplasty (Pascoe, 1979a). The competence of the vaginal seal can be assessed via the use of a sterile probe, or may require a speculum. Speculum examination of the vagina and cervix is discussed in detail in the following section.

The selection of a mare with a Caslick vulvoplasty for breeding must be made with considerable caution. There are a finite number of times that the operation can be performed and, unless the more complicated Pouret operation (Pouret, 1982) is resorted to, there is, by implication, a limit to the mare's breeding career. An additional consideration is one of equine welfare and whether such poorly conformed mares should be bred, as, in so doing, the trait is perpetuated within the equine population.

11.2.6. Internal examination of the reproductive tract

Examination of the internal reproductive tract of the mare is a skilled veterinary surgeon's job, necessitating the use of several examination techniques. Information given by these assessments can be indispensable in assessing the reproductive potential of a mare.

11.2.6.1. Vulva and vagina

Vaginal internal assessment can be carried out by means of a speculum (Fig. 11.4). The most commonly used is a Caslick's speculum (Fig. 11.5).

The speculum consists of either an expandable or a non-expandable hollow metal tube, with a light source attached. The sterilized and well-lubricated speculum is inserted into the mare's vagina. If appropriate, this is inserted in the closed position and is expanded, slowly opening up the vagina.

Fig. 11.4. Vaginal-speculum examination of the mare's vagina. The independent light source allows the inside of the vagina to be illuminated and hence easily viewed.



Fig. 11.5. A non-expandable vaginal speculum.

The light source then illuminates the mucous-membrane lining and the cervix, allowing examination in detail. Disposable speculums of plastic or coated cardboard are also available, but these need an independent light source and so the use of two hands, making techniques such as swabbing very difficult. The use of a vaginal speculum must be accompanied by several precautions as, when in position, it opens up both the vulval and the vaginal seals. The procedure should be carried out under conditions as sterile as possible, in a dust-free environment. The mare's tail should be bandaged and the perineal area thoroughly washed (Fig. 11.6).

A more sophisticated technique, termed an endoscopy, can be used today for viewing the mare's vagina and cervix and ultimately the rest of the internal reproductive tract. Older endoscopes were rigid and therefore had limitations to their use; the newer, flexible, fibre-optic endoscopes are more versatile. As in humans, the endoscope can be used to view any internal structures, by viewing either

through the body wall and into the body cavity or via the oral/nasal cavity to the digestive/respiratory system, via the rectum into the large intestine or, finally, via the vagina to view the reproductive tract.

The endoscope consists of a series of flexible carbon-fibre filaments with a light source and camera attached. The flexible carbon-fibre rod is passed through the vagina and up into the inner reproductive tract. One set of the carbon-fibre filaments allows the passage of light from the external light source down the endoscope, illuminating the internal structures. The other set allows the transmission of the image back to the camera and on to a television monitor. The positioning and angle of view of the endoscope can be controlled remotely from the exterior end of the endoscope (Fig. 11.7).



Fig. 11.6. Use of a vaginal speculum in the mare.



Fig. 11.7. Endoscopic examination of the reproductive tract of the mare. The attendant on the right controls the optic-fibre camera, while the attendant on the left guides the endoscope within the mare's reproductive tract. The image produced is viewed on the TV monitor. (From Thoroughbred Breeders Equine Fertility Unit, Newmarket.)

The use of the endoscope is expensive and is usually confined to examination of the upper, less accessible parts of the reproductive tract (Fig. 11.7; Le Blanc, 1993a; Threlfall and Carleton, 1996).

In the healthy mare, both the speculum and the endoscope should show the mucous membranes lining the vulva and vagina to be a healthy pink colour. Any mucous present should be clear and not cloudy or yellow-white in colour. An infected vagina appears red and inflamed and is possibly covered in a cloudy mucus. Infections of the vagina are not uncommon and will be discussed in the following section on infections and also in Chapter 19.

The vulva and vagina should be free from bruising, irritation, scars and tears. They are susceptible to damage and injury at parturition and so they should be carefully checked. Adhesions, if present, are caused by scar tissue and may render the mare unserviceable or cause her pain at mating and hence rejection of the stallion. AI is a possible alternative but she may still have problems at foaling.

Neoplasms of the vulva and vagina are relatively rare, though, if present, may cause problems at covering. Three kinds of neoplasms can be seen in horses: first, melanomas, malignant growths of the melanin-containing pigment cells, particularly prevalent in grey mares and not necessarily confined to the vulva and vagina; second, carcinomas or malignant neoplasms of the epithelial cells; and, finally, papillomas or benign neoplasms. If a mare does show evidence of neoplasms, it is debatable whether she should be mated, as there is evidence to suggest that the tendency to develop some neoplasms is heritable.

Finally, urovagina (pooling of urine within the floor of the vagina) may be observed. Stagnant urine held within the vagina will increase the chance of infection. Urine pooling is evident in mares with poor perineal conformation and in older multiparous mares in which vaginal tone has been lost. It may also result from past vaginal damage or injury.

11.2.6.2. *Cervix*

The cervix may be considered as the connection and final seal between the outer reproductive tract and the inner, more susceptible tract. As such, its competence as a seal is very important. This may be assessed by means of a speculum, as described above for vaginal examination. The cervix varies greatly with the stage of the oestrous cycle and pregnancy and, as such, can aid in diagnosis. During oestrus, the cervix is fairly relaxed, pink in colour, its tone flaccid and any secretions quite thin, and it appears to 'flower' (relax) into the vagina. This is to ease the passage of the penis at copulation. During dioestrus and, to a greater extent, during anoestrus, the cervical tone increases, it appears whiter in colour, the seal becomes tighter and the secretions become thicker. During pregnancy, the cervix is again tightly sealed and white in colour, with a mucous plug acting to enhance the effectiveness of the seal. The tone of the cervix may also be examined via rectal palpation. In general, the state of the cervix should correspond to ovarian activity and that of the rest of the tract. If not, infections or abnormalities should be suspected. Infection of the cervical mucosa, or cervicitis, will be discussed in detail later (Chapter 19). Cervicitis is characterized by a red/purple swollen cervix, often protruding into the vagina and covered with mucus, clearly seen at speculum examination. In severe cases, cysts may be evident and may result in adhesions and long-term permanent damage, preventing correct closure of the cervical seal, with resultant infection. Adhesions as a result of cervical damage may close it completely, preventing entry of the penis and so the passage of sperm to the upper tract and the draining of fluid from the uterus, resulting in serious uterine infections. Minor cervical damage and/or adhesions can be helped by surgery.

11.2.6.3. *Uterus*

Examination of the mare's uterus is a more complicated procedure, as the uterus forms part of the inner and less accessible reproductive tract. An appreciation of its gross structure

can be obtained by rectal palpation and/or ultrasonic scanning. The colour and condition of the mucous membranes of the uterus may be inspected by means of an endoscope and further examined by uterine biopsy.

Rectal palpation is a commonly used and relatively effective way of obtaining a tactile impression of the inner reproductive tract and so identifying structural abnormalities (Figs 11.8 and 11.9). The procedure involves restraining the mare in stocks and inserting a well-lubricated, gloved hand and arm through the anus and into the rectum of the mare. The wall of the rectum is fairly thin, so the reproductive tract, which lies

immediately below it, can be palpated through the rectum wall. The procedure, however, must only be carried out by an experienced operator, as rupturing the rectum wall is not uncommon. Rectal palpation can be used to assess the tone, size and texture of the uterus, uterine horns, Fallopian tubes and ovaries. In addition, cysts, tumours, neoplasms, stretched broad ligaments, uterine endometritis, sacculations, adhesions, lacerations, scars and delayed involution can be identified. As such, it is a useful, cheap and immediate aid to selection of mares (Greenhof and Kenney, 1975; Shideler, 1993c).

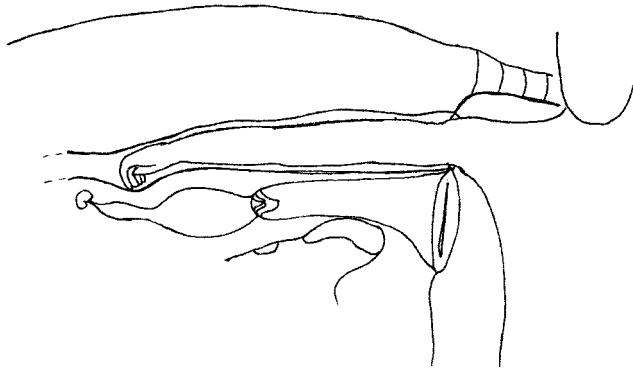


Fig. 11.8. Rectal palpation in the mare.



Fig. 11.9. Rectal palpation in the mare allows the practitioner to feel the reproductive tract through the relatively thin wall of the rectum.

Ultrasonic scanning is a newer and more expensive method of assessment, which gives an immediate, visual impression of the upper reproductive tract, rather than the tactile interpretation obtained with rectal palpation (Jones, 1995; Sertich, 1998). The ultrasonic scanner is based on the Doppler principle. When high-frequency sound waves hit an object, they are absorbed or deflected back to a varying extent, depending on the density of that object. The reflected sound waves are transduced to appear as an image on a visual display unit. Solid objects appear white and fluid appears black, with many variations of grey in between (Fig. 11.10; Toal, 1996). In the case of ultrasonic scanners used in the assessment of a mare's reproductive status the transducer or ultrasonic emitting and receiving device can be inserted into the rectum or less commonly placed on the mare's flank and directed towards the reproductive tract (Fig. 11.11; Ginther and Pierson, 1983). The resultant image can be used to identify abnormalities similar to those assessed via rectal palpation but, in particular, the presence of cysts, intrauterine fluid, air, debris,

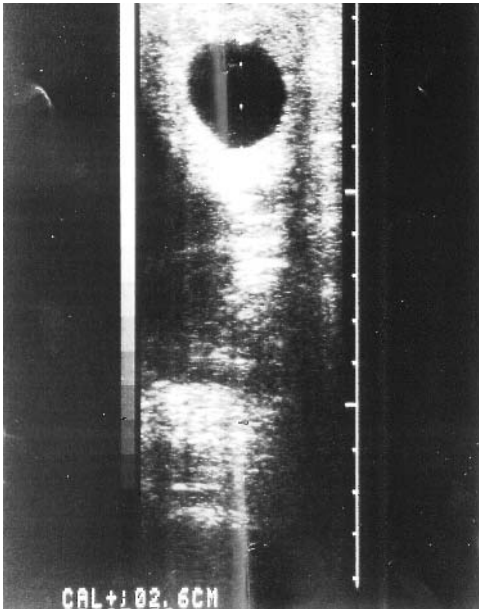


Fig. 11.10. The image produced by an ultrasonic scanner; fluid areas show up as black and solid structures as white, with variations in between.

neoplasms, etc. (Ginther, 1984; McKinnon, 1987a,b,c, 1998). The technique can also be used to assess the size and shape of the uterus and ovaries and so can be used to determine ovarian activity (Adams *et al.*, 1987; Perkins, 1996).

By means of rectal palpation and ultrasonic scanning, the healthy uterus should appear either flaccid with little tone if the mare is in oestrus, or turgid with plenty of tone if she is in dioestrus. If the palpater works his/her way gradually from either uterine horn and across the uterine body, local thickenings, cysts or fibrotic areas can be detected and the diameter of each horn estimated and compared. The two horns should match. Mares that have had a history of infections may show ventral out-pushings of the uterus, especially at the junction between the uterine body and horns. These mares may well have difficulty in carrying another pregnancy to term, as they often lose the fetus at the time of implantation. Incomplete involution of the uterus post-partum can also be detected, indicating infection, weakening or rupture of the uterine wall, and, in severe cases, the uterus may never completely involute. This leaves a flaccid area of uterus with inadequate tone compared with the remainder. Again, such mares may well have difficulty carrying a pregnancy to term. Haemorrhages within the broad ligaments may be identified as local hard swellings up to 40 mm in diameter. Stretched broad ligaments may also be detected, often due to excessive strain, accidents or damage. Again, such mares may be incapable of carrying a pregnancy to term, the broad ligaments being unable to support the weight of a full-term fetus.

An endoscope, as described earlier in this chapter, may be used to examine the internal surfaces of the reproductive tract and to give a real-time image. Once the endoscope is inserted, sterile air or oxygen is passed into the uterus, easing the viewing of the mucous membranes and uterine epithelium (Fig. 11.7; Mather *et al.*, 1979; Le Blanc, 1993b). The normal healthy uterine epithelium is pale pink in colour and any mucus

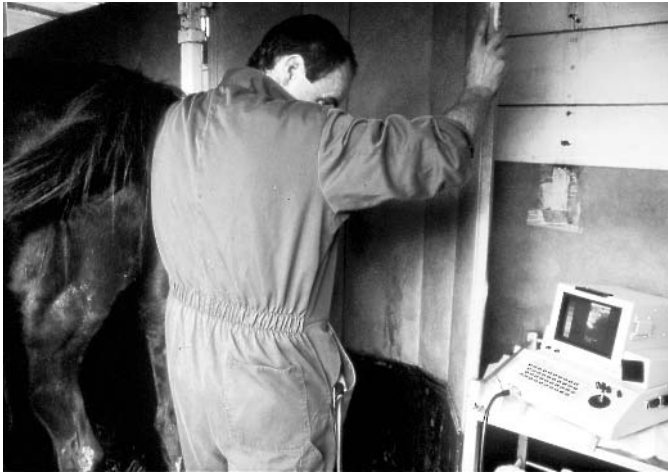


Fig. 11.11. The use of an ultrasonic scanning machine in the mare.

present is clear. Any signs of redness or discoloration within the uterus, especially if it is associated with cloudy or creamy mucus, are indicative of uterine infection – endometritis. Evidence of cysts, blood clots, thickening of the endometrium or scar tissue may indicate possible future problems in the mare's reproductive life.

The use of the endoscope is as yet limited, due to the cost of the instrument and its delicate nature. Endoscopes at present are normally restricted to research use and large specialist veterinary practices.

If any areas of concern within the mare's uterus are detected by rectal palpation, ultrasound or endoscopy, then a uterine biopsy may be performed to aid further investigation. Uterine biopsies are taken during endoscopy and involve the removal

of a small section of uterine endometrium by means of a small pair of forceps (Figs 11.12 and 11.13). The biopsy forceps are passed into the vagina and guided up through the cervix by a well-lubricated, gloved hand. The index finger guides the forceps through the cervix and into the uterus. The gloved hand is then removed and inserted into the rectum to guide the forceps to the section of the uterus that is to be biopsied. The jaws of the forceps are opened and the section of endometrium to be sampled is pushed between the jaws by this hand. The jaws are then shut and the forceps drawn back slowly until the pressure of the endometrium is felt. A sharp tug will then release the sample, which is immediately removed and fixed in readiness for histological examination.

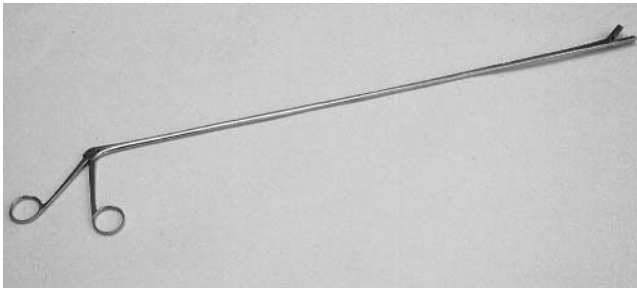


Fig. 11.12. Uterine biopsy forceps.

Fig. 11.13. The use of uterine biopsy forceps in the mare. The arm inserted via the rectum is used to ease the wall of the uterus into the jaws of the biopsy forceps.

Samples taken from the mid-uterine horn have been shown to be representative of the uterine endometrium as a whole (Bergman and Kenney, 1975), so they are suitable for routine biopsies where no specific abnormality is being investigated. In biopsies to further investigate suspected abnormalities, samples should be taken from each area in question and another sample from a seemingly normal area of endometrium. Biopsies can be taken at any time of the oestrous cycle, but normally standard biopsies are taken in mid-dioestrus in order to standardize results (Kenney, 1978; Doig and Waelchi, 1993).

Such samples can allow identification of abnormalities, as well as indicating evidence of uterine degeneration. The examiner looks for changes in cell structure, especially luminal cells, which may be diagnostic of inflammation, fibrosis or necrosis (Doig and Waelchi, 1993). The process is reported not to be disadvantageous to the mare's reproductive cycle, though some people report a delay in the next oestrous period (Kenney, 1977).

Not all the methods discussed are used as standard selection criteria, but they may be used to elucidate any indications of abnormalities picked up by the simpler, more routine procedures of rectal palpation and ultrasonic scanning.

11.2.6.4. Fallopian tubes

The competence of the Fallopian tubes is more difficult to assess. As mentioned previously, uniformity in size and shape can be assessed via rectal palpation. In such an examination, the Fallopian tubes should feel wiry and uniform in consistency when rolled between the palpater's fingers. Severe abnormalities may be detected in this way, especially adhesions connecting the infundibulum to the uterus or ovaries and scar tissue. However, salpingitis (inflammation of the Fallopian tubes) is hard to detect. Though it has been reported to be relatively uncommon in mares, any such inflammation is of importance, as it may hinder the passage of ova along the Fallopian tube and may even result in complete blockage of the oviduct.

Assessment of Fallopian-tube blockage is very difficult. The traditional starch-grain test is relatively successful but complicated to carry out and therefore of limited practical use. A starch-grain solution is injected through the mare's back at the subluminal fossa, using a long needle. The ovary is manipulated, via rectal palpation, to lie immediately beneath the needle. Starch grains (approximately 5 ml) are then injected on to the surface of the ovary. Washings are collected from the anterior vagina and cervix 24 h later. The presence of starch grains in the flushings indicates that the Fallopian

tube associated with the ovary on to which the starch grains were injected is patent.

A more invasive form of investigation is the use of laparotomy under general anaesthesia. This technique involves the exteriorization of the uterine horns and Fallopian tubes through an incision made either in the mare's flank or in the abdomen. The Fallopian tubes and ovaries can then be examined in detail. Such techniques are expensive and highly invasive; they also run the normal risks associated with general anaesthesia and are therefore rarely used.

Laparoscopy, involving the insertion of a rigid endoscope with a light source into the body cavity via the abdominal wall, is a further possibility, allowing the upper tract to be directly viewed *in situ*. This technique is slightly less invasive but does not allow such detailed examination (Fig. 11.14). Its expense and the risks of general anaesthesia again make it a rare specialist technique.

11.2.6.5. Ovaries

Ovarian activity can be assessed most simply by rectal palpation and more exten-

sively by ultrasonic scanning. The appearance of the mare's ovaries varies considerably with season and reproductive activity. These changes are detailed and discussed in Chapter 1. Rectal palpation has traditionally been used to assess the stage of the mare's oestrous cycle and therefore help in the timing of mating. It can be used in this context of selection criteria to ensure that the mare is reproductively active and that follicles and corpora lutea (CL) are being produced. It can also be used to ensure that the reproductive stage shown within the ovaries is synchronized with developmental changes in the remainder of the mare's tract. Such techniques can also indicate the presence of adhesions and neoplasms, as well as cystic follicles, ova fossa cysts and other ovarian abnormalities, which may disrupt the mare's reproductive activity. Further detail on the assessment of the reproductive stage of the mare by ovarian examination is given in Chapter 13.

Laparoscopy has been used experimentally to elucidate ovarian problems. Its use in practical stud-farm management is as yet limited, due to cost and the need for general anaesthesia (Fig. 11.14).

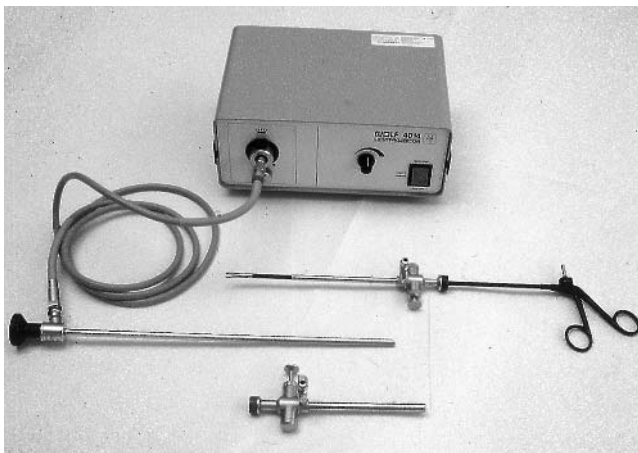


Fig. 11.14. A laparoscope, which may be used to view the internal structures of the mare's reproductive tract via the abdominal cavity. The trocars pictured in the foreground are inserted through puncture wounds in the mare's abdomen. Through the larger is passed the laparoscope for viewing the internal organs, and through the smaller is passed the manipulating forceps for moving internal organs.

11.2.7. Infections

Mares are notorious for being susceptible to reproductive-tract infections, in particular endometritis (Ricketts and Mackintosh, 1987). Not only can these cause temporary and possibly permanent infertility, but they can also be transferred relatively easily to the stallion and on to other mares. It is imperative, therefore, that infected mares are identified so that they can be discarded in the selection process and/or treated. One of the standard procedures used to detect such infections is swabbing. Swabs may be taken from the uterus, cervix, urethral opening and/or clitoral area and assessed for pathogenic organisms – for example, venereal disease (VD) bacteria *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Taylorella equigenitalis*, which are true sexually transmitted bacteria, or *Streptococcus zooepidemicus*, *Escherichia coli* or *Staphylococcus*, which are not strictly VD bacteria but which may also be transferred at covering and can cause uterine infections.

Uterine swabs should only be taken during oestrus when the cervix is relaxed and moist, easing the swab's passage, and at the time when the mare's natural immunological response to accidentally introduced bacteria is heightened due to the dominance of oestrogen. The swabs may be guided in through the cervix using a speculum or guided via a gloved, lubricated arm, as in the case of uter-

ine biopsies. Once the swab is in the lumen of the uterus, it is rotated against the endometrium to absorb uterine secretions and bacteria. The risk of accidental contamination of the swab by either airborne pathogens or bacteria present in the cervix and/or vagina is minimized by careful washing of the mare and the use of a guarded swab. Such swabs are contained within two sterile tubes. The first tube is passed through the cervix and the second tube is telescoped into the uterus. The swab can then be pushed out through the second tube far enough to reach the endometrium. A reversal of this process will reduce the contamination on retraction of the swab (Figs 11.15 and 11.16; Greenhof and Kenney, 1975).

Cervical and urethral-opening swabs are easier to obtain and may be collected at any time of the cycle. The samples may be taken using a vaginoscope by a similar process to that described for the uterus. The swab absorbs the secretions of the cervix itself.

Clitoral swabs are taken from the clitoral sinuses around the clitoral fossa. Gentle squeezing of the clitoris may produce smegma secretions for swabbing; care, however, should be taken, as some mares object. The clitoral fossa harbours, among other pathogens, *Taylorella equigenitalis*, the causal agent for contagious equine metritis.

Once the sample has been taken, the swab can be applied to a variety of growth mediums, incubated under various condi-

Fig. 11.15. Cervical swabbing in the mare.

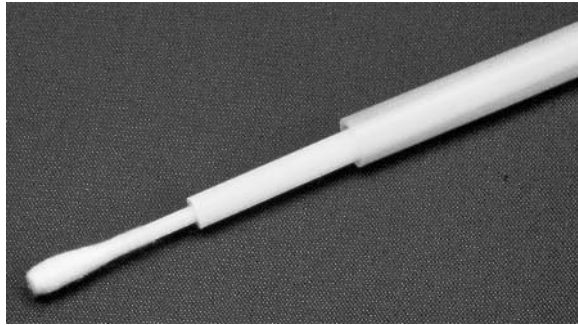


Fig. 11.16. A guarded swab used for bacterial assessment of parts of the mare's reproductive tract.

tions and subsequently examined for specific bacterial colonies. Incubation under different temperature, humidity, atmospheric pressure and oxygen content aids the identification of specific pathogenic types. Alternatively, swabs can be smeared on to glass slides for cytological examination (Ricketts *et al.*, 1993).

Swabbing is a very effective method of assessing bacterial contamination of the mare's reproductive tract, but results must be treated with some caution. The presence of bacteria within the vagina and cervix does not necessarily indicate uterine infection, especially if the cervical seal is fully competent. The natural flora of many mares contains pathogenic organisms. The actual process of taking a sample also increases the chance of reproductive-tract contamination by airborne bacteria; hence the technique must be carried out under as clean and sterile conditions as possible. In general, therefore, as indicated above, swabbing is best carried out during oestrus.

Providing the possible drawbacks are borne in mind, swabbing is a simple procedure to carry out in the routine selection of mares for breeding. Indeed, as a result of the Thoroughbred Breeders Association *Annual Code of Practice* (1978 onwards), it is advised that all Thoroughbred mares and many of those in other breed societies should be swabbed prior to arrival at the stud and again at the oestrus of service, to ensure the absence of VD bacteria. Only mares with a negative certificate will be accepted at the stud and only those with a second negative certificate will be covered.

11.2.8. Blood sampling

If there are reasons to believe that a mare may have problems in carrying a foal to term and no anatomical abnormalities have been detected using the previously discussed techniques, then the problem may lie in hormonal inadequacies. The endocrine profiles of mares can be determined by sequential blood sampling. Any deviations from the normal profile can be identified and the specific area of failure, e.g. follicle development, ovulation signal, oestrous behaviour or CL regression, can be identified. In the light of these results, appropriate hormone therapy may be possible to compensate for the natural deficiencies, or it can be decided that such a mare is not worth the risk or cost of such therapy. Details on the normal endocrine profiles for mares are given in Chapter 3.

Blood samples can also be used to indicate the general health status of a mare, bringing to light specific deficiencies related to diet, low-grade infections, blood loss, cancer and parasite burdens. Low red-cell counts, i.e. below $10 \times 10^6 \text{ ml}^{-1}$, indicate anaemia. Packed-cell volume is a quick and easy assessment of red blood cell:fluid balance: normal levels are 40–50%. Assessment of the colour of the supernatant in the packed-cell-volume test is a useful problem indicator. Normally strawlike in colour, discolouration can indicate problems; for example, red/pink indicates a breakdown of red cells and release of haemoglobin. Haemoglobin levels themselves are a well-established indicator of anaemia, levels $<12\text{--}17 \text{ g l}^{-1}$ indicating problems.

A high white blood-cell count, above 12,000–14,000 ml⁻¹, indicates the presence of disease – in particular, infection or cancer. A total protein index (protein content of serum after clotting) of <15 g l⁻¹ indicates blood loss, starvation or liver, kidney or gastrointestinal disease. Fibrinogen levels may also be indicative of abnormalities; high levels, >10 g l⁻¹, suggest inflammatory, neoplastic or traumatic disease (Table 11.1; Varner *et al.*, 1991; Pickett, 1993c).

11.2.9. Chromosomal abnormalities

Blood samples may also be used for chromosomal analysis, if genetic abnormalities are suspected (Bowling *et al.*, 1987). Genetic abnormalities reported include a missing X chromosome, Turner's syndrome (63XO), chimerism or mosaic (63XO:64XX), sex reversal (64XY) and extra X chromosome (65XXX) (Hughes *et al.*, 1975; Halnan, 1985; Bowling, 1996).

11.3. The Stallion

The selection criteria for reproductive competence in the stallion are similar to those of the mare and can be listed as follows:

- History
- Temperament and libido
- Age
- General conformation
- Reproductive tract examination
- Semen analysis
- Blood sampling
- Infections
- General stud management

Reproductive evaluations are necessary prior to purchase but may also be used routinely prior to each breeding season or if a problem is suspected (Thompson, 1994).

11.3.1. History

Records of a stallion's history are invaluable in aiding selection and, as with the mare, can be divided into his breeding and general history. Records for stallions tend not to be as detailed or as readily available as those for mares.

11.3.1.1. Reproductive history

Records of his past breeding performance, if available, should answer questions such as: When does his season normally start and end? How many mares is he used to covering in a season? What are his return rates like? What is his semen quality like?

The answers to these questions will indicate his reproductive ability. Stallions with short seasons will be less able to cover as many mares and may suffer from low libido. The number of mares he has served per season in the past and the return rates, along with semen analysis, will give an indication of what workload he will be capable of. If his return rates are high, especially if a significant decrease is seen with an increase in workload, this may indicate the natural limit of the number of mares he is able to cover. The routine of covering may affect performance and can be tailored to suit the stallion. Routines may involve one or two covers per day for 6 days with a day's rest, two covers per day for 8 days followed by 2 days' rest or numerous variations on these themes. Most

Table 11.1. Blood parameters indicative of disease.

Parameter	Value indicative of disease
Red blood-cell count	< 10 × 10 ⁶ ml ⁻¹
Packed-cell volume	< 40–50%
White blood-cell count	> 12,000–14,000 ml ⁻¹
Total protein index	< 15 g l ⁻¹
Haemoglobin	< 12–17 g l ⁻¹
Fibrinogen	> 10 g l ⁻¹

stallions do need a rest day but should be able to cover mares at the rough frequency of the systems given above (Pickett *et al.*, 1985). If there are indications that a stallion is not capable of such workloads and requires more rest days to maintain his fertility rates, then his selection should be queried, especially if you are looking for a stallion to purchase. Return rates are a good guide to a stallion's ability (Van Buiten *et al.*, 1999), but it must be remembered that the fertility of a stallion is only as good as the fertility of the mares he is covering.

Any previous semen analysis should also be detailed in his records. Many Thoroughbreds have a semen analysis carried out routinely at the beginning of each season. This, along with a blood sample, normally taken at the same time, allows any potential problems to be identified in time for remedial action to be taken before the breeding season starts. Any past reproductive-tract infections should also be detailed in a stallion's records, along with any treatment given and the outcome. Any long-term effects of infection should be evident in the stallion's workload and return rates for the rest of that season and any subsequent seasons.

11.3.1.2. General history

The stallion's general history should indicate his vaccination and worming status, along with the incidence of injuries and accidents. Damage to his hindquarters or limbs may restrict his ability to mount a mare, as may laminitis and neurological disorders (Griffin, 2000). AI may be an alternative (Davies Morel, 1999); even so, he is likely to need the occasional mount for the collection of semen samples, though the number of mounts per mare fertilized will be significantly reduced and the unpredictability of mounting a mare avoided. Such stallions are not advised for purchase. As with the mare, if there were any suggestions that any damage or weaknesses might be heritable, selection would not be advised. Injuries to a stallion's genitalia, usually as a result of a kick from a mare, will cause degenerative and scar tissue within the testes, which will reduce the

number of germinal and Sertoli cells and hence the fertility rates of the stallion. Severe damage resulting in the removal of a testicle should also be noted in a stallion's records to reassure potential purchasers that he is not a rig. Such stallions are capable of fertilizing a mare, but the workload may have to be reduced. Severe injuries to a stallion during mating often have psychological effects, drastically reducing his libido, possibly to such an extent that he is unwilling to cover naturally.

Past illnesses should also be indicated in his records. Illnesses associated with the respiratory or circulatory systems may indicate that the stallion will not be capable of working a full season, limiting the numbers of nominations available or that can be sold. Again, if there is a possibility that such weaknesses could be heritable, the stallion should be avoided. Any illnesses resulting in a fever can disrupt spermatogenesis, due to the elevated temperature (Johnson *et al.*, 1997). This may result in temporary infertility, though this may not be evident for several months as the spermatogenic cycle takes 57 days (Davies Morel, 1999). Systemic infections, for example, strangles or flu, can cause inflammation within the testis and, if this results in a significant amount of tissue degeneration, permanent subfertility or even infertility may result.

The sorts of stallion records that should be available to a potential purchaser are illustrated in Fig. 11.17.

11.3.2. Temperament and libido

The temperament of the stallion is very important, for ease of management and as a heritable trait. A stallion of a quiet and kind disposition is a great asset and will be much easier and safer to handle (Fig. 11.18). A stallion that is rough to his mares will not only run the risk of inflicting permanent damage on them but may also be hurt himself if they retaliate. A rough stallion will prove unpopular and it may be difficult to get him enough mares to make his use economic. Some protection, in the form of neck guards,

Date of mare's arrival	Mare/owner	1st service date	2nd service date	Return date	Return date	Positive PD	Live foal d.o.b.	Comments
16.1.00	Trefaes Fancy Mr T. James	20.3.00	22.3.00	—	—	4.4.00	22.2.01 filly	Foal at foot, d.o.b. 12.2.00
18.1.00	Katie Jane M. Davies	15.3.00	16.3.00	7.4.00	—	21.4.00	Abortion 10.12.00	Maiden mare
30.3.00	Penpontpren Mina Mrs M. Morel	30.3.00	1.5.00	21.4.00	22.4.00	15.5.00	15.3.01 colt	Barren mare
1.2.00	Lluest Megan Lluest Stud	30.4.00	1.5.00	21.5.00	—	—	—	Foal at foot, d.o.b. 30.3.00 Delayed uterine involution
12.2.00	Rose Mr A. Jones	18.3.00	—	—	—	8.4.00 twins aborted	—	Foal at foot, d.o.b. 12.3.00 Twins detected 8.4.00
12.2.00	Lady Jane Barnswood Stud	19.3.00	20.3.00 21.3.00	—	—	5.4.00 twins, one pinched	2.3.01	Barren, twins detected 5.4.00, smallest pinched

Fig. 11.17. The type of records that should be kept for each stallion during his breeding season, indicating the mares he has been put to and the result, along with any noteworthy comments.



Fig. 11.18. A well-behaved stallion is an asset to any stud, easing his management and reducing the danger to his handlers.

can be given to mares that are mated to stallions that tend to bite during covering, but no protection can be given against stallions that are downright vicious and, as such, they should be avoided at all costs.

Ideally, records should indicate the stallion's temperament and any specific characteristics that he might have. It is to be hoped that his bad habits, especially those that might prove dangerous, will also be indi-

cated. To be forewarned is to be forearmed and might lead you to reject an unsatisfactory stallion.

Bad behaviour in many stallions is a direct result of the conditions and management under which they are kept (Chapter 18). Therefore, especially in the case of a stallion that seems to have developed bad habits later in life or after a change of owner or management, the conditions under which he is kept should be assessed before he is rejected for covering a mare. However, as a potential purchase, he is not a good choice, as such habits are difficult to break. Bad behaviour tends to perpetuate itself, as, due to the potential danger, such stallions are kept confined for longer periods of time and hence away from other companions. Their boredom is exacerbated and their bad habits develop further. Stereotypes (habits) to be aware of include: weaving; crib-biting; and wind-sucking; all signs of boredom. Additionally, there is a commonly held belief, though not supported by scientific research, that other horses may copy stereotypes.

Stereotypes such as self-masturbation were once frowned upon but are now considered natural behaviour, of no consequence except the potential embarrassment to owners. Some stallions also indulge in self-mutilation, especially after mating, biting themselves in areas where the smell of the mare lingers. Though thorough washing post mating can reduce the incidence, the potential for self-harm and the added management time and expense may preclude their selection.

A stallion's reproductive temperament and willingness to cover are termed his libido, which partly determines his reproductive potential. Libido is governed, like all other sexual activity, by season (Chapter 4). Hence, those stallions with longer seasons tend to show a higher libido and therefore willingness to mate early on in the season, extending the time in which he can be worked. Ideally, if selecting a stallion to purchase, he should be seen teasing and covering a mare. A stallion with a low libido will need to mount a mare several times before ejaculation, taking up to 20 min to cover a

mare, or he may fail completely; he may also show initial interest very reluctantly. The number of mounts per ejaculation and the time between actual intromission and ejaculation are good indications of libido. The number of mounts per ejaculation should be as near to one as possible and the time between intromission and ejaculation a matter of seconds (Thompson, 1994).

11.3.3. Age

The age of the stallion is less important than that of the mare, as far as reproductive ability is concerned. The significance of age in the selection of the stallion depends on what that stallion is required for, i.e. for a single mating to a selected mare or as a potential purchase for long-term future use. If you are selecting him for service of a single mare, then, as far as you are concerned, he will be required to perform on just a couple of occasions; his age is of limited importance, providing he is capable of covering. However, if you are looking to select a stallion for purchase and therefore long-term future use, you want to ensure that he is young and fit enough to give you plenty of seasons, but old enough to have proved his worth.

As far as a lower limit is concerned, most colts reach puberty at 18–24 months (see Chapter 4; Clay and Clay, 1992). A colt can, in theory, be used as soon as he reaches puberty, but care must be taken to introduce him to the job gradually and not to overwork him too soon or give him awkward mares, which may affect his as yet delicate ego and reproductive confidence (Johnson *et al.*, 1991). Further details on early stallion management are given in Chapter 18. The purchase and use of such young stallions are risky, as they have no proven performance record.

As far as an upper age limit is concerned, this really depends on the stallion's general health and condition. If he has no problems with lameness, stamina, wind, injury, etc., he may well be capable of working well into his teens and even twenties, though in the latter years his workload may have to be reduced. There is reported evidence that reproductive

capability is inherently reduced with age (Johnson and Thompson, 1983; Amann, 1993a,b). However, other work disputes this, suggesting that any decline in reproductive performance with old age is indirect, due to reducing libido from injury, arthritic conditions, etc. and not a decline in spermatogenesis *per se* (Johnson, 1991a).

As discussed in the case of mare selection, if an older ex-performance horse is being considered, it must be borne in mind that he will require a prolonged period of time to adjust physically and psychologically to his new role in life. Details of the problems associated with using performance horses as stallions are given in Chapter 12.

11.3.4. General conformation and condition

A stallion's general conformation is of importance, not only as it will be passed on to his offspring, but also to ensure that he is capable of withstanding a full breeding season. A stallion of poor limb conformation, especially the hindquarters, will also be weak in this area and may therefore be unable to withstand the heavy workload of a full season, limiting his economic viability.

Particular note should be taken of his physical ability to cover mares. He should be free of all signs of lameness, especially in the hind limbs. His legs should be checked before and after exercise and a comparison made to ensure that there is no sign of swelling, a sign of possible weakness. He should be free of all conditions such as arthritis, spinal or limb injury, wobbler syndrome or laminitis, all of which could cause pain. A stallion's feet should also be in excellent condition, regularly trimmed to ensure they stay that way. Adequate heart room in a broad chest is also desirable and, if doubt is placed on the stallion's cardiovascular system, electrocardiography may be conducted.

Good general condition and physical fitness are very important for the breeding stallion. The condition of a stallion, like that of the mare, can be classified on a scale of 0–5

(0 emaciated, 5 obese). The optimum body condition for a stallion in work is 3, that is, he is well muscled up and in fit working condition (Fig. 11.19). Stallions in condition score less than 3 tend to have lower libido and are physically less able to stand a heavy workload (Jainudeen and Hafez, 1993). If the stallion's condition is very poor, spermatogenesis may also suffer. At the other extreme, obese stallions also tend to have low libido; they tend to be lazy and may be incapable of mounting a mare. In addition, the extra weight puts additional strain on the mare at mating and may cause her damage. It is to be remembered that the nutritional demands during the breeding season are similar to those of a performance horse, the workload of the two being approximately equivalent (Thompson, 1994; Griffin, 2000). Further details on stallion nutritional management are given in Chapter 18.

11.3.5. External examination of the reproductive tract

An external examination of the stallion's reproductive genitalia is an essential selection procedure, as his ability to perform is naturally a function of the condition of his



Fig. 11.19. A stallion in good, fit, well-muscled working condition ready for the breeding season.

reproductive organs (Griffin, 2000). He should have two normally functioning testes, which may be felt through the scrotum and palpated to ensure they are of a similar size and consistency, move easily within their tunicae and are not warm to the touch. Occasionally, the left testis is slightly larger than the right, but the difference is slight and should not be accompanied by an increase in heat. The surface of the testis should feel smooth, with the occasional blood vessel being felt running under the skin. Any adhesions preventing the testes from moving up and down easily within the tunica are likely to indicate scar or fibrous tissue due to past injuries. This not only reduces the volume of functioning testicular tissue but may also interfere with spermatogenesis within the remaining tissue. Indeed, testicular size is a very good indicator of the spermatozoa-producing capacity of the stallion and hence his potential workload. As such, testicular volume has been advocated as an assessment criterion when selecting for reproductive potential (Love *et al.*, 1991; Pickett and Shiner, 1994). Excessive fat within the scrotum as a result of excessive body condition will also increase the insulation of the testes, with the danger of increasing testicular temperature and therefore decreasing sperm production.

Malignant or benign growths within the testes are rare but may be evident (Caron *et al.*, 1985; Schumacher and Varner, 1993). The skin of the scrotum should be checked for dermatitis, which can cause an increase in testicular temperature. The position of the epididymis should also be felt. Their normal position in the non-retracted relaxed testis is on the cranial (abdominal) side of the scrotum. Positioning elsewhere may indicate testes torsion or twist (Hurtgen, 1987; Threlfall *et al.*, 1990).

The vas deferens leaving the testis, plus testicular blood and nerve supply, passes up into the body of the stallion through the inguinal canal, which should be free from adhesions and hernias.

The penis and prepuce of the stallion should also be examined for injury, squamous-cell carcinoma, summer sores, sarcoids

and general infections or injury. Examination can be carried out at washing prior to semen collection and should be a routine selection procedure. Details on VD infections and penile conditions are given in Chapter 19.

11.3.6. Internal examination of the reproductive tract

As with the mare, examination of the internal reproductive tract of the stallion is a skilled veterinary surgeon's job. Information given by internal examination can be indispensable in assessing the reproductive potential of a stallion, though internal examination may be limited by financial implications and the need for experienced personnel.

Access to the internal parts of the stallion's reproductive tract is very difficult. Some appreciation may be gained by rectal palpation and ultrasound (Little, 1998). Via rectal palpation, the vas deferens can be felt entering the body cavity at the inguinal ring and both, one either side, should feel smooth and of uniform diameter. Alongside the vas deferens as they enter the body cavity lies the spermatic artery, the pulse of which should also be checked. Very low blood pressure, or a drop between successive examinations, may be indicative of a haemorrhage, blood clot or tumour or the release of body fluids into a localized infection site. The accessory sex glands may also be palpated individually and their texture, size and shape assessed. Paired glands, such as the seminal vesicles, should be checked for symmetry. Ultrasound may be used to give an indication of physical abnormalities and accessory-gland secretory function (Weber and Woods, 1993). An indication of the function of the accessory glands may also be gained by semen evaluation, as will be discussed in the following section.

11.3.7. Semen evaluation

Semen evaluation is a routine selection procedure. If a stallion is to cover mares throughout the breeding season with consistent

success, his semen has to meet various minimum parameters. In many studs, all stallions routinely have their semen evaluated at the beginning of each season and if a problem is suspected. The quality of his semen has a direct effect on the stallion's ability to consistently and successfully cover a number of mares throughout the season (Jasko *et al.*, 1990a,b, 1991; Gastel *et al.*, 1991; Pickett, 1993a; Parlevliet and Colenbrander, 1999). Ideally, for an accurate evaluation, samples should be taken either: (i) one after 3 days' sexual rest preceded by a double collection taken 2 h and 1 h prior to test collection; (ii) as the last collection of a series of seven daily collections, preceded by a double collection taken 2 h and 1 h prior to test collection; or (iii) both collections taken 1 h apart after 1 month's sexual rest (Pickett and Voss, 1972; Kenney, 1975a; Sullivan and Pickett, 1975; Swierstra *et al.*, 1975). In most commercial enterprises/AI programmes, such regimes are not economically viable, and single sampling, interpreted with caution, can provide adequate information for most routine practices.

Collection of semen is normally by means of an artificial vagina. Details of the collection and evaluation procedures are given in Chapter 20 and elsewhere (Davies Morel, 1999). The normal parameters for semen are given in Table 20.3.

11.3.8. Infections

Like the mare, the stallion is susceptible to sexually transmitted diseases and, as such, all stallions should be tested for infections prior to purchase, either to eliminate them or to allow treatment to commence prior to their use.

As with the mare, swabs can identify infections of the genital tract; these are taken from the urethra, the urethral fossa and the prepuce of the stallion's penis. Swabs should be taken from the erect penis, erection being encouraged by an oestrous mare or tranquilizers. Three different swabs must be used and it is best to take the urethral-fossa sample last, as this one can cause considerable discomfort and hence objection. Swabs or cultures of semen samples can also be

assessed. The stallion's semen and penis have a natural microflora of bacteria and fungi and these should be distinguished from VD pathogens. The most noteworthy bacteria, classified as VD causers of acute endometritis, are *K. pneumoniae*, *P. aeruginosa* and *T. equigenitalis* (Couto and Hughes, 1993; Parlevliet *et al.*, 1997).

Swabbing is routinely carried out in many studs on all their stallions well before the season starts. This allows time, if infections are identified, for treatment to begin and take effect before the breeding season. Further details of infection of the stallion's reproductive tract and the effect upon reproduction are given in Chapter 19.

11.3.9. Blood sampling

Blood sampling of stallions can be used to assess their general health and can indicate low-grade infection, blood loss, cancer, nutritional deficiencies or parasite burdens. Details on the information that can be gathered from blood sampling have been given in the previous section on the selection of the mare. Any stallion showing these characteristics should not be considered for use until the problem has been identified and appropriate treatment commenced.

Blood samples are rarely used for hormone analysis, since the considerable inter-stallion variation reduces the accuracy of such testing to assess potential reproductive performance (Roser, 1995). The episodic nature of testosterone release also necessitates a period of sequential blood sampling from which an average can be taken rather than a single representative sample. Low plasma testosterone concentrations have been associated with low libido and poor semen quality (Watson, 1997).

11.3.10. Chromosomal abnormalities

Chromosomal abnormalities are well documented in the mare but less so in the stallion (Long, 1988). However, conditions such as XX male syndrome (64XX), chimerism or mosaic

(64XX:64XY), Klinefelter's syndrome (65XXY) and 13 quarter/deletion (64XY) are associated with an inability to impregnate mares or very low fertility rates, despite apparently normal genitalia (Halnan and Watson, 1982; Halnan *et al.*, 1982; Bowling *et al.*, 1987; Bowling, 1996).

11.3.11. General stud management

If your selection of a stallion is not for purchase but rather for use on one of your mares, you will also be interested in the management at the stud at which he stands. There are several things that will concern most owners selecting a stud to send their mare to, and these will be discussed in turn.

The system of breeding used is of prime importance. Is the stud appropriately equipped for visiting mares or are you expected to bring the mare for the day, having detected yourself that she is in oestrus, and take her away the same day after covering? Some studs allow the mares to stay a few nights but have only limited facilities and may well expect mares to live out. This obviously has a bearing on the distance it is possible to travel. You should also consider the method of covering, varying from pasture breeding to intensive in-hand breeding. The various methods used are discussed in detail in Chapter 13. Some studs will expect the mare to be taken home as soon as she has been covered; others will allow her to stay for re-covering if necessary and will only allow her home after a positive pregnancy diagnosis at scanning and/or rectal palpation, usually 2–4 weeks post-mating.

Many of the smaller native-pony-type studs do not have the facilities to foal down visiting mares, necessitating mares to be brought to stud very soon after foaling. This can be traumatic and dangerous for the foal and precludes using a stud that is too far away. Larger studs tend to have the mares brought in to foal, normally 4–6 weeks prior to foaling. This allows the mare to be covered on her foal heat without the danger of transporting a young foal.

The daily management at the stud should also be investigated and matched as closely

as possible to the mare's normal routine. If not, her routine at home should be slowly altered to that at the stud to minimize the stress of change. All animals on the stud should be wormed regularly and vaccinated, and documented proof of adequate protection is usually required of all visiting mares. At the more intensive studs, mares standing to valuable stallions will also require negative certificates to a variety of VD bacteria (Horse Race Betting Levy Board, 2001).

A good impression of the standard of management of a yard can be gained by a general visit. The yard, whatever system in use, should be clean and tidy, all the mares and stallions should be in good condition, the pasture well tendered and the animals contented. If the mare is to foal there, the foaling facilities should be clean, safe and roomy, with a good system for 24 h monitoring by skilled staff. The facilities of the yard and the equipment and expertise available will reflect the type of stallion and his nomination fee.

The system that you choose is ultimately a personal choice, depending on your priorities and the finances available. The more intensive systems tend to be associated with the Thoroughbred industry, where expense is of less concern but hygiene and protection of valuable stock are of paramount importance. In such intensive systems, mares are taken to the stud to foal, are subsequently covered and possibly re-covered and remain at stud until pregnancy is confirmed, often at days 12 and 25. In such systems, the service fees are high and the costs of keep and veterinary attention are great, but this is offset by the value of the offspring and the risks are lower. At the other end of the spectrum, native studs will serve a mare that arrives in their yard and within half an hour she can be on her way home. In such systems, stallion fees are low, as are costs, but the offspring is often of low value and the risks are higher.

Further details on the management systems and principles for both the mare and stallion at breeding are given in the following chapters, Chapters 12–19. When examining potential studs it is as well to bear in mind that the ideal is not normally achieved. It is unrealistic to expect a yard standing a cheaper

stallion, with stud fees of £50–£100, to have the facilities found in a Thoroughbred stud standing stallions with nomination fees up to £50,000.

11.4. Conclusion

If more attention were paid to assessment and selection of breeding stock on the basis

of reproductive competence, significant amounts of money, time and effort would be saved in trying to breed from subfertile or infertile stock. Failure to select fertile stock not only leads to suffering for the mare and stallion, but also frustration for all. Additionally, many problems are inherited and so will be perpetuated in subsequent generations, to the detriment of the equine population as a whole.

12

Preparation of the Mare and Stallion for Covering

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12.1. Introduction

It is essential that preparation of the mare and stallion starts in plenty of time prior to covering. It is of no use making a last-minute decision that you wish to put a mare in foal or stand a stallion at stud and then wonder-

ing why she does not hold or your return rates are high. A preparation time of at least 6 months is required in order to maximize the chance of conception. This chapter will concentrate mainly on this period, with some reference to any earlier preparation that may be required.

12.2. Preparation of the Barren and Maiden Mare

If a mare is destined to be a brood mare, then she must be brought up with this aim in mind, with close monitoring of her general condition and growth. Horses normally reach puberty at 1.5–3 years of age, depending on the breed and their nutritional status in early life. If you are considering breeding a mare at or near puberty, before she has reached her full mature size, then her stage of development and her general condition are of utmost importance. Breeding before attainment of mature body size places on the mare the additional burden of pregnancy as well as that of her own continued growth and development. Providing she is well grown and in good body condition, she should be able to cope with such additional demands, but this must be borne in mind when considering her management through pregnancy, especially that of nutrition.

Today, many horses, both mares and stallions, are bred for the first time relatively late in life, having already had a successful performance career, on the basis of which they have been chosen for breeding. In such cases, both mares and stallions are older and more set in their ways and have been managed to date as athletes, not breeding stock (Fig. 12.1).

Performance horses must be allowed plenty of time to unwind both physically and psychologically. This should start during the autumn prior to the planned covering, during which time workloads should be slowly reduced to a maintenance level. This is usually adequate for most horses, though much variation is evident between individuals, and some may take as long as 18 months to adjust. Intensive training is detrimental to reproduction in all mammals; this can be clearly demonstrated in women athletes, who regularly fail to ovulate. Mares in peak athletic condition will characteristically demonstrate abnormal oestrous cycles, often showing delayed oestrus, silent heats and oestrous behaviour not accompanied by ovulation. However, given a long enough adjustment period, a mare should start showing regular oestrous cycles and can be successfully mated.

Careful attention should also be given to the mare's nutrition and, related to that, her exercise. Mares should be in good, not fat, body condition at covering. The body-condition score to aim for at mating is 3 (on a scale 0–5). In addition, as indicated in Chapter 11, it is generally believed that in maiden and barren mares better conception rates are obtained if they are on a rising plane of nutrition, in particular increasing energy, in the last 4–6 weeks prior to covering; this is termed flush-



Fig. 12.1. Many successful stallions are currently or have been performance horses. As such, they need careful management in order to perform well at both jobs.

ing (Van Niekerk and Van Heerden, 1972; Kubiak *et al.*, 1987; Morris *et al.*, 1987). Flushing of barren mares has also been reported to advance the start of the breeding season by as much as 30 days (Fig. 12.2; J. Newcombe, Wales, 2001, personal communication). The best regime is to ensure that the mare is in condition score 2–3 in the autumn prior to covering. As the season approaches, her energy intake can be gradually increased by replacing some of her roughage intake with concentrates in the last 4–6 weeks (Hintz, 1993a; Guerin and Wang, 1994; Frape, 1998).

If the mare is young, it may also be pertinent to supplement her diet during this period with protein, calcium, phosphorus and vitamin A. Requirements for these elements are higher in young maiden mares than in mature mares.

Exercise is also important, helping to maintain body condition and prevent obesity. Gentle riding or hacking out provides a good form of exercise for barren mares during the preparation period (Fig. 12.3). All mares should be turned out daily and ideally barren mares, especially those not broken,

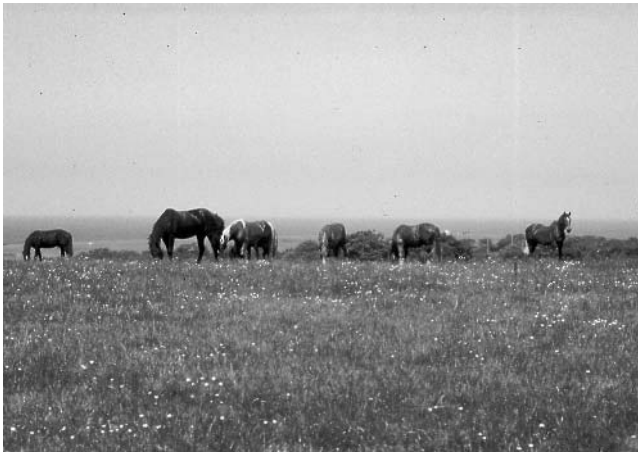


Fig. 12.2. Mares being flushed on lush pasture prior to mating in order to encourage optimum fertilization rates.



Fig. 12.3. Gentle riding provides ideal exercise for barren mares, in order to keep them fit for covering and any resultant pregnancy.

would live out except in the most inclement weather, thus providing them with *ad libitum* exercise (Fig. 12.4).

A period of 6 months' preparation also allows time for a mare to be tested for infection, the appropriate treatment to be given and full recovery to occur. Any damage due to old infections can be investigated and either assisted in its repair or the mare discarded from the breeding scheme. If she is of sufficient genetic merit, the possibility of embryo transfer (ET) can be investigated and necessary arrangements made. All barren mares, especially maidens, should be introduced to new handling systems, buildings and surroundings associated with breeding during this period. This is of particular importance with ex-performance mares, which are usually moved or sold in readiness for their new career. Familiarization with management practices associated with breeding, such as restraint in stocks, rectal palpation, ultrasonic scanning, teasing, etc., should also be ensured prior to covering. Old-timers need only to be reintroduced a few weeks before the season starts. Any changes in diet must be introduced gradually and also the introduction of new companions should be done early enough to allow a settling down period. Providing all changes are made gradually and in good

time, stress at covering can be minimized, so maximizing reproductive success.

12.3. Preparation of the Pregnant Mare

Care must also be taken in the preparation of the pregnant mare for re-covering the following spring. However, the presence of a pregnancy limits to a large extent how she can be managed. One advantage with a pregnant mare is that she has seen it all before, at least in the previous year, and no psychological and physiological adjustment is required. However, a careful eye should be kept on her condition in order to prevent obesity. Once a pregnant mare is overweight, it is very difficult to rectify, especially in late pregnancy, without endangering the fetus. Prevention is therefore much better than cure and it is essential that the mare is in a fit condition prior to initial covering and that condition 3 is maintained throughout pregnancy and into her next covering. Flushing of pregnant mares is not advised and has no effect or possibly a detrimental effect on conception rates to the foal heat (Frape, 1998). If there is a reasonable period of time between foaling and re-covering, this can be used to try and adjust body condition, but care must



Fig. 12.4. If mares are not broken to ride, they must be turned out to provide enough exercise in order to keep fit (Penpontbren Welsh Cob Stud).

be taken, as alteration in nutrition will affect milk yield and hence the foal at foot. There is conflicting evidence with regard to the effect of nutrition during late pregnancy and between parturition and foal heat or subsequent conception rates (Henneke *et al.*, 1981; Jordan, 1982). However, it is generally accepted that a condition score of 3 is ideal during this period and that mares should be fed well in lactation to stimulate fertility (Frape, 1998).

Exercise is an important aid in maintaining a body condition of 3 in the pregnant mare. Pregnant mares can normally be safely ridden up to the sixth month of pregnancy; but this depends on the individual. By the sixth month, all strenuous work must be excluded. Mares that are not broken and those in late pregnancy should be turned out every day to help maintain fitness and blood circulation and prevent boredom. In an ideal world, mares would live out with plenty of opportunity to exercise: such systems are popular in temperate latitudes without the risk of adverse weather. Exercise not only helps to prevent obesity but also maintains the mare's fitness and muscle tone, both of which will be needed at and after parturition.

12.4. Preparation of the Stallion

If a colt is destined to become a working stallion, he must be brought up during his early life with this aim in mind, especially with regard to discipline. Many stallions become hard to handle and, in some cases, downright dangerous, because discipline and respect for authority have not been established in early life.

Most of the problems encountered in stallions previously used as performance horses are behavioural abnormalities. They will have had several years during which they will have been actively discouraged from displaying any sexual behaviour. As a result, they may be severely inhibited at their first sight of an oestrous mare, anticipating punishment. They will often find it hard to revert to natural stallion behaviour and need varying amounts of time to adjust to their new

career. Many stallions take a few seasons to completely adjust and some never really do achieve complete adjustment. A stallion's libido may also be affected, and such stallions may, as a result, always prove to be slow to react to an oestrous mare and show clumsy mounting behaviour.

As well as their psychological adjustment, attention should be paid to a stallion's nutrition and exercise during this preparation period. He must be fit, not fat. A heavy covering season places significant demands on the stallion, especially in terms of energy, and he will often lose condition over the season. This loss in condition is minimized if the stallion's energy intake is increased by increasing the concentrate proportion of his diet and if he is fit and in a body condition score 3 as the season commences (Fig. 12.5). Both excess and low body weight reduces a stallion's libido. Exercise helps a stallion maintain good condition, preventing obesity and maintaining muscle tone and stamina. Stallions have a tendency to become obese, as they are regularly kept individually in stables or paddocks away from each other and mares. In natural conditions, they would of course be free to exercise at will.

If stallions are badly behaved, it is tempting to keep them confined, with only limited turnout. This perpetuates the problem and accentuates any misbehaviour due to boredom. Some stallions can be safely ridden, providing an excellent form of exercise, as well as good discipline (Fig. 12.6). In the less intensive studs, usually where stock is less valuable, some quiet stallions are turned out in July at the end of the season with either their mothers, an old mare or other quiet mares. They can then be brought back into riding work and possibly hunted over the winter. This system allows a rest period after the season, followed by a fitness regime prior to the start of the next season. It also provides them with another purpose in life, which greatly helps discipline.

At least four weeks prior to his first mare, the stallion should be brought into the stud environment. He should then be introduced or reintroduced to the yard, handling systems, buildings, surroundings and especially the



Fig. 12.5. A stallion should be in a fit, not fat, condition – that is, condition score 3 at the beginning of the breeding season (Penpontbren Welsh Cob Stud).



Fig. 12.6. Riding provides an excellent form of exercise, as well as good discipline.

covering area in plenty of time to allow familiarization prior to the first covering. Any changes in diet should be introduced slowly, before the season starts, along with any new companions. Further details regarding stallion management are given in Chapter 18.

12.5. General Aspects of Reproduction

Many performance horses have been on various drug regimes during their performance

lifetime. Plenty of time must be allowed for the body to eliminate these drugs from the system. Corticosteroids, used as anti-inflammatory drugs for the treatment of various injuries, can have serious detrimental effects on reproductive performance in both stallions and mares. They can also impair the body's ability to fight infection, which is of particular significance in the mare. The use of anabolic steroids to boost muscle development and hence performance also has severe detrimental effects on reproductive performance and these should be eliminated from

the animal's system well in advance of the breeding season (Shoemaker *et al.*, 1989).

Many breed societies recommend and, in some cases, require that all mares and stallions should be swabbed prior to covering. Infections of the reproductive tract are a major cause of infertility. Many studs, especially in the Thoroughbred industry, require all mares being covered to hold a current negative certificate to a number of venereal-disease bacteria and viruses (*Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Taylorella equigenitalis*, equine herpesvirus, equine viral arteritis) prior to arriving at the stud. Swabbing to identify bacteria may also be required again at the oestrus of covering. Stallions may also be required to be swabbed to test for venereal diseases prior to each season and also during the season if infection is suspected (Crowhurst *et al.*, 1979; Ricketts *et al.*, 1993; Horse Race Betting Levy Board, 2001). Further details on venereal diseases are given in Chapter 19.

12.6. Manipulation of the Oestrous Cycle in the Mare

As the majority of equine matings are today influenced by humans, we have therefore, attempted to control the timing of covering to our advantage. There are several methods by which the timing of mating can be manipulated: first, by altering the beginning of the season; and, second, by manipulating the timing of the mare's oestrus and ovulation within that season.

12.6.1. Advancing the breeding season

The horse is classified as a long-day breeder, with a breeding season extending on average from April to November in the northern hemisphere and October to May in the southern hemisphere (Chapter 3). The Thoroughbred industry and, increasingly, other breed societies register the birth of all foals on 1 January (northern hemisphere) or 1 July (southern hemisphere), regardless of their actual birth date. It is therefore desir-

able, in order to achieve maximum advantage in the racing industry, to ensure that mares foal as soon after 1 January as possible. As the mare has an 11-month gestation, mares need to be covered at the beginning of February. Hence the arbitrary covering season in Thoroughbreds in the northern hemisphere runs from 15 February to 1 July and in the southern hemisphere from 15 August to 31 December. Other breed societies also have similar arbitrary breeding seasons. Even in societies that do not stick to set breeding seasons, it is an advantage to have foals born as early as possible in the year in order to maximize their size during the showing and event season and enhance their chances of success.

The existence of an arbitrary breeding season that does not correspond to the natural one is a major limiting factor in breeding mares. Manipulation of the mare's breeding cycle is required to advance the timing of oestrus and ovulation in order to accommodate these artificial limits. There are several means by which the breeding season in the mare may be advanced. These include the use of light and hormone therapy, or a combination of the two.

12.6.1.1. Light treatment

In a population of intensively managed mares, only 10% will voluntarily show oestrus and ovulation during the non-breeding season. This figure is much lower for extensively managed feral populations. Artificial manipulation of light, along with nutrition and temperature, will significantly increase this percentage. Of these three environmental factors, manipulation of light is the most successful (Meyers, 1997).

Light treatment of mares to advance the breeding season was first pioneered by Burkhardt (1947), with later work by Kooistra and Ginther (1975) among others. Mares can be introduced to a 16 h light:8 h dark regime either suddenly or gradually. Light can be delivered by means of a 200-watt incandescent source per 4 m × 4 m loose box, or equivalent. A slightly lower-watt light source may be used if the stables

are lined by reflective material. Light treatment can be started any time from early November onwards, but it is essential that the mare experiences an initial autumnal reduction in day length prior to light manipulation. Commencement of light treatment early in December will result in coat loss within 4 weeks, followed by ovarian activity normally 2–4 weeks later. The season may be advanced by up to 3 months, but there is considerable individual variation (Kooistra and Loy, 1968; Kooistra and Ginther, 1975; Oxender *et al.*, 1977; J. Newcombe, Wales, 2001, personal communication). In general, the earlier in the year light treatment begins, the longer the time interval to oestrus and ovulation. Light treatment from early December is often chosen, as it results in ovarian activity during early February. More recent work suggests that a window of 1 h light 8 h after dusk would suffice, resulting in the same effect as 16 h light (Scraba and Ginther, 1985). However, in practice, as the timing of dusk varies, it is easier to administer a 16 h block of light.

The effects of light manipulation may be enhanced by increases in ambient temperature and nutritional levels, both of which are also associated with spring. However, the relatively minor effect that temperature and nutrition have on advancing the season mean that their use beyond that of rugging up mares and increasing their energy intake is not justified (Allen, 1978; Meyers, 1997). Although light treatment is successful in advancing the season, the timing of response in a group of mares is very variable. In an attempt to reduce this variation between mares, hormone treatment was investigated.

12.6.1.2. Exogenous hormonal treatment

The use of exogenous hormones to induce out-of-season breeding is very successful in anoestrous ewes and, as such, is a useful management tool. Unfortunately, the anoestrous mare's ovary appears relatively insensitive to exogenous hormone therapy. Treatment with pregnant mare serum gonadotrophin, human chorionic gonadotrophin (hCG) or follicle-stimulating hormone

(FSH) by single or multiple injection, as used in other livestock, results in little if any response. However, natural crude equine pituitary extract, if injected daily over a period of 2 weeks, does induce oestrus out of season (Douglas *et al.*, 1974; Lapin and Ginther, 1977). Such a long period of treatment is required as the mare's ovary apparently requires a prolonged period of gonadotrophin stimulation in order to adequately develop follicles so that they can react to an ovulating agent (see Chapter 3). Short-term treatments will not develop follicles to an adequate stage to allow them to react to any ovulation stimulus.

The use of gonadotrophin-releasing hormone (GnRH), naturally responsible for inducing FSH and luteinizing hormone (LH) production by the pituitary, has also been investigated. Both GnRH infusion, via mini pumps or subcutaneous implants, and injection over a period of time have been reported to give some success (Hyland *et al.*, 1987; Johnson 1987a; Hyland and Jeffcote, 1988; Palmer and Quelier, 1988; Ainsworth and Hyland, 1991; Harrison *et al.*, 1991; Mumford *et al.*, 1994). Further development of the use of GnRH involved its inclusion in a hormone regime in an attempt to mimic the natural concentrations of FSH, LH and progesterone in the normal oestrous cycle. A single injection of GnRH is followed by daily injections of progesterone for 8 days. This regime is repeated twice more to give three artificial cycles. This treatment, though not effective in mares in deep anoestrus, is reasonably successful in the transitional stage from anoestrus to the breeding season (Evans and Irvine 1976, 1977, 1979).

12.6.1.2.1. PROGESTERONE. Progesterone has been used to induce oestrus and ovulation in anoestrous mares, but it also seems only to be effective in the transition period. It is known that it is the decline in progesterone prior to ovulation that encourages LH and FSH release, so finally maturing and ovulating follicles. Without this progesterone decline, the reaction in terms of the concentration of LH and FSH released is limited and therefore ovulation does not occur or it

occurs in the absence of oestrus. The resulting corpus luteum (CL) of any such ovulation is often incompetent, and only after the second ovulation of the season are acceptable conception rates achieved. Therefore, if progesterone is artificially administered and then withdrawn, it mimics the natural decline in progesterone and helps induce the normal increase in LH and FSH required for ovulation and results in a competent CL (Freedman *et al.*, 1979). Ovulation accompanied by oestrus will occur within 7–10 days after the cessation of a 10–12-day progesterone treatment period.

Progesterone is commonly used in the management of breeding mares. Traditionally, it was administered as a progestagen (Altrenogest) either orally (Regumate) or via intramuscular injection. More recently, it has been available for administration via a progesterone-releasing intravaginal device (PRID) (Rutten *et al.*, 1986), a controlled internal drug-releasing device (CIDR) (Lubbeke *et al.*, 1994), sponges (Dinger *et al.*, 1981) or Cu-mate, all of which are impregnated with progesterone, rather than progestagen, and placed within the mare's vagina for the duration of the treatment. Oestrus and ovulation after removal of PRID, CIDR and Cu-mate are reported to be quicker than after Regumate or injection, by virtue of the fact that the progesterone within PRID, CIDR and Cu-mate allows folliculogenesis to continue. Additionally, the resulting elevated systemic progesterone levels decline over the later part of the treatment period, as the finite amount of progesterone within the devices is absorbed, further ensuring significant follicle development prior to their removal. The interval from the end of treatment to oestrus and ovulation is reported to be 2–4 days for PRID and CIDR, compared with 8–10 days for Regumate (Squires, 1993a,c; Arbeiter *et al.*, 1994; Newcombe and Wilson, 1997; J. Newcombe, Wales, 2002, personal communication). Additionally, progestagen appears to depress follicular activity, which can only resume after the end of treatment, hence resulting in a longer interval to oestrus and ovulation. Intravaginal sponges, PRID and CIDR have the advantage of being labour-saving, being required only to be

inserted and removed. They also ensure that a standard dose is administered. However, they can be lost and can cause vaginitis, which, though of no real consequence to conception, looks unsightly. In common with these, the advantage of injection is that you can be assured that each mare has received her allotted dose. However, it is more expensive, especially when you consider the costs of the veterinary attendance, now required by law in the UK for the administration of injections to horses. Oral administration has the advantage that no vet is required, but some mares may refuse to take the feed in which it is mixed. If only part of the food is eaten, it is impossible to know how much progestagen has been taken. Furthermore, all mares have to be fed individually. While considering the use of progesterone, it is worth noting that progesterone use has been associated with a decrease in neutrophil production in response to a bacterial challenge. This may well be significant in mares with poor perineal conformation or a history of uterine infections.

12.6.1.2.2. PROSTAGLANDINS. A further alternative for use in late anoestrous mares is prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), administered in a series of two or three injections at 48 h intervals. $PGF_{2\alpha}$ is luteolytic in nature – that is, it destroys the CL and terminates the luteal phase of the cycle. In the natural cycle, in the absence of a pregnancy, prostaglandin is produced at a specific time (12–14 days) after ovulation. As such, it both marks and causes the termination of the luteal phase and the commencement of the endogenous hormone changes associated with oestrus and ovulation (Allen and Rowson, 1973; Loy *et al.*, 1979; Neely *et al.*, 1979; Cooper, 1981; Savage and Liptrap, 1987; see Chapter 3). It is this association with the termination of one cycle and commencement of the next that is exploited in this treatment. Ovulation has been reported in 73% of mares treated in this way (Jochle *et al.*, 1987).

Prostaglandin use does have some side-effects, largely relating to the ability of prostaglandin to activate smooth muscle. Its use may therefore be linked to increased

gastrointestinal activity, manifested as diarrhoea, sweating and possibly slight caudal ataxia (Le Blanc, 1995). These side effects vary with the analogue used and the individual mare; however, providing the recommended dose rate is not exceeded, they are not serious.

12.6.1.3. Combination treatments

A combination of light treatment and hormone therapy has been used and gives encouraging results. As indicated previously, many hormone treatments, though more successful at timing oestrus and ovulation than light treatment, only work when used during the transition phase. Light therapy can therefore be used to advance mares into the transition period; exogenous hormones can then be used to time oestrus and ovulation more precisely. Progesterone alone plus light treatment can be successful. Light treatment (16 h light and 8 h dark) should be introduced during November/December (or May/June in the southern hemisphere), followed by progesterone for a period of 10–15 days from early to mid-January (July) onwards (Palmer, 1979; Squires *et al.*, 1979a; Heeseman *et al.*, 1980; Scheffrahn *et al.*, 1980). The exact timing of the progesterone treatment depends on when covering is planned and when light treatment is commenced.

Some of the best and most consistent results have been obtained using a similar regime but with the addition of $\text{PGF}_{2\alpha}$: 16 h light and 8 h dark for 6–8 weeks, followed by 10 days' progesterone treatment; $\text{PGF}_{2\alpha}$ is

then administered on day 10 to induce luteolysis of any naturally occurring CL. It is reported that up to 82% of mares ovulated within 9–16 days of treatment and achieved a conception rate of 65% (Bristol, 1986). Table 12.1 gives an example of such a regime.

Though there are very many ways, in theory, of advancing the breeding season in the mare, most are too expensive and time-consuming to make them commercially viable and they often achieve unreliable results. In practice, most commercial studs just use light treatment, possibly supplemented by progesterone.

12.6.2. Synchronization of oestrus

In addition to advancing the breeding season, there are many other reasons for manipulating the timing of oestrus – mainly to ease mare management. In many countries of the world, e.g. South America and the USA, mares are run in large herds that roam over vast tracts of land. In such systems, handling needs to be kept to a minimum. It would therefore be ideal if mares could all be treated in batches right through from conception to birth and foal rearing. In order for this to be successful, a reliable and exact method of timing ovulation and oestrus is required. Such treatment also alleviates the need for teasing, rectal palpation, scanning, etc. and would be most useful in conjunction with artificial insemination (AI). Such treatment would also allow the ovulation and

Table 12.1. The advancement of oestrus and ovulation in the mare, using light treatment and progesterone supplementation. AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0 (15 Dec)	Light treatment commenced 16 h light : 8 h dark
Day 28 (12 Jan)	Coat loss in mare may be apparent
Day 42 (26 Jan)	Ovarian activity may be apparent
Day 43 (27 Jan)	Progesterone treatment started
Day 55 (8 Feb)	Progesterone treatment stopped
Day 60+ (13 Feb)	Oestrus commences
Day 62+ (15 Feb)	Ovulation may occur – covering/AI

oestrus of a single mare to be timed, again to ease her management but specifically useful for AI and ET.

12.6.2.1. Progesterone

Progesterone supplementation and subsequent withdrawal may be used to time oestrus and ovulation. The use of progesterone or one of its analogues works on the principle of imitating the mare's natural dioestrous or luteal phase. This is achieved by mimicking the natural progesterone production through the administration of exogenous progestagens. Termination of this artificial luteal state, achieved by the cessation of treatment, acts like the end of the natural luteal phase and so induces the changes in the mare's endogenous hormones responsible for oestrus and ovulation (Bristol, 1993).

Within 2–3 days of progesterone supplementation, a mare will normally cease all oestrous activity, which will remain suppressed until treatment is terminated (Loy and Swann, 1966). After 15 days' treatment, oestrous behaviour is apparent at 3–7 days and ovulation at 8–15 days after progesterone withdrawal (Table 12.2; Van Niekerk *et al.*, 1973; Squires *et al.*, 1983a). Conception rates are comparable to those associated with naturally occurring oestrus (Van Niekerk *et al.*, 1973; Squires *et al.*, 1979a, 1983a).

Traditionally, long periods of progesterone supplementation were used, up to 20 days: though oestrus was suppressed, ovulation did occasionally occur during treatment. Hence, the timing of ovulation was not that successful (Loy and Swann, 1966).

Shorter periods of progesterone supplementation are now being used. As the period of progesterone supplementation may not be long enough to ensure that the natural CL has regressed, a combination of progesterone supplementation and prostaglandin treatment is used. This is discussed in the following section.

12.6.2.2. Prostaglandins

PGF_{2α} (Prostin F2 Alpha and Dinoprostomethamine 5) or one of its analogues (Alfa prostol fluprostenol, prostalene or frenprostalene) provides a successful means of timing oestrus and ovulation in the mare (Bristol, 1987; Le Blanc, 1995). As discussed previously, PGF_{2α} both marks and causes the termination of the luteal phase and the commencement of the endogenous hormone changes associated with oestrus and ovulation (Allen and Rowson, 1973; Loy *et al.*, 1979; Neely *et al.*, 1979; Cooper, 1981; Savage and Liptrap, 1987; Bristol, 1993; see Chapter 3). As such, administration of exogenous prostaglandins, providing it is within certain time limits within the luteal phase, allows its termination to be controlled and, with it, the timing of oestrus and ovulation.

The success of prostaglandin in timing oestrus in the mare is variable and depends upon the stage of the cycle. The CL of most mares is refractory to prostaglandin treatment prior to day 5 (Douglas and Ginther, 1972; Loy *et al.*, 1979). A good response is normally obtained when treatment is given between days 6 and 9 (Loy *et al.*, 1979). To be

Table 12.2. The timing of oestrus and ovulation in the mare, using progesterone supplementation. PRID, progesterone-releasing intravaginal device; i.m., intramuscular; AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0–16	Progesterone supplementation (intravaginal sponges or PRID, i.m. injection or oral administration)
Day 19 onwards	Oestrus
Day 21 onwards	Ovulation may occur – covering/AI

successful, the treatment must not only terminate the luteal phase but also induce ovulation. Considerable variation exists between the time of prostaglandin treatment and ovulation; a range of 24 h to 10 days is reported (Loy *et al.*, 1979). The time interval is determined by the stage of follicular development at treatment. Follicles at 4 cm in diameter or greater ovulate on average within 6 days, though again considerable variation is reported. If the follicle ovulates within 72 h, it is often accompanied by an abbreviated oestrus or no oestrus at all. Occasionally, when a large follicle is present, prostaglandin treatment results in the regression of that follicle and the development and subsequent ovulation of another follicle; hence there is a longer time interval between treatment and ovulation (longer than 8 days) (Loy *et al.*, 1979). The most consistent results are obtained when treating mares earlier in the cycle, with small follicles (less than 4 cm in diameter) as less variation exists and the interval to ovulation is, on average, 6 days (Table 12.3).

The previously described use of prostaglandin relies upon a single injection, the major disadvantage of which is that the stage of the mare's oestrous cycle must be

known. In smaller intensive studs, where individual mares are monitored, this may present no problems. However, in large groups of mares, kept in herd situations, or in mares whose stage of the oestrous cycle is unknown, a double injection of prostaglandin is required (Hyland and Bristol, 1979). These two prostaglandin injections need to be administered 14–18 days apart (Table 12.4).

The timing of the onset of oestrus with such a treatment is quite successful; 60% of mares are reported to commence oestrus within 4 days of the second injection and 92% show oestrus within 6 days (Hyland and Bristol, 1979; Voss *et al.*, 1979; Squires, *et al.*, 1981a; Squires, 1993a,c). Nevertheless, the synchrony and timing of ovulation are still very variable. Ovulation may occur anywhere between 2 and 12 days after the second injection.

12.6.2.3. Human chorionic gonadotrophin

Further refinement of these protocols includes the use of hCG, a human placental gonadotrophin with LH- and FSH-like properties. As such, it enhances and supplements the natural release of gonadotrophins, which

Table 12.3. The timing of oestrus and ovulation in the mare, using a single injection of prostaglandin. AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0	Oestrus
Day 7	Prostaglandin
Day 9	Oestrus commences
Day 11	Ovulation may occur – covering/AI

Table 12.4. The timing of oestrus and ovulation in the mare, using two injections of prostaglandin. AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0	Prostaglandin
Day 16	Prostaglandin
Day 20	Oestrus commences
Day 22	Ovulation may occur – covering/AI

drive follicular development and, more specifically, ovulation. Its use is advocated to hasten ovulation and reduce the duration of oestrus (Table 12.5; Voss *et al.*, 1975; Harrison *et al.*, 1991).

Several timings for the injection of hCG have been advocated, most of them between 4 and 6 days after the second prostaglandin injection (Douglas and Ginther, 1972; Palmer and Jousset, 1975; Hyland and Bristol, 1979; Voss *et al.*, 1979; Bristol, 1981; Squires *et al.*, 1981a). Palmer and Jousset (1975) reported that 75.8% of mares ovulated within 72 h of an hCG injection, which was given 6 days after the second prostaglandin injection. Yurdaydin *et al.* (1993) achieved similar success using hCG on day 5 post prostaglandin. When used on day 8 after the PGF_{2α} injection, oestrous synchronization rates of 90% have been reported (Holtan *et al.*, 1977). Other work, however, has demonstrated a more variable response or no significant improvement with the use of hCG (Holtan

et al., 1977; Squires *et al.*, 1981a). It has been advocated that hCG be used twice, on day 7 (7 days after the first PGF_{2α}) and on day 21 (7 days after the second PGF_{2α} injection) (Table 12.6). The aim of this is to encourage the development of a competent CL from the first prostaglandin injection, which would then react with less variation to the second prostaglandin injection. This regime is reported to result in up to 95% of mares ovulating on either day 22 or 23 (Allen *et al.*, 1974; Palmer and Jousset, 1975; Voss, 1993).

Though reasonably successful in inducing ovulation, hCG has a major disadvantage. With repeated administration mares are reported to develop antibodies to and therefore may become refractory to hCG (Roger *et al.*, 1979; Wilson *et al.*, 1990), though this effect is not reported to be a problem by some (J. Newcombe, Wales, 2001, personal communication). GnRH and its analogues have been advocated for use in its place.

Table 12.5. The timing of oestrus and ovulation in the mare, using two injections of prostaglandin and a single injection of human chorionic gonadotrophin (hCG). AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0	Prostaglandin
Day 15	Prostaglandin
Day 19	Oestrus commences
Day 20	hCG
Day 21	Ovulation may occur – covering/AI

Table 12.6. The timing of oestrus and ovulation in the mare, using two injections of prostaglandin and two injections of human chorionic gonadotrophin (hCG). AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0	Prostaglandin
Day 7	hCG
Day 14	Prostaglandin
Day 18	Oestrus commences
Day 21	hCG
Day 22	Ovulation may occur – covering/AI

12.6.2.4. Gonadotrophin-releasing hormone

GnRH acts to stimulate the natural release of LH and FSH from the anterior pituitary. As such, its administration as a series of multiple injections (four at 12 h intervals) or via a subcutaneous implant has been demonstrated to significantly advance the onset of ovulation in mares with follicles greater than 3 cm in diameter (Table 12.7).

Success rates of 88–100% of mares ovulating within 48 h of treatment (with deslorelin) have been reported (Johnson, 1986b; Harrison *et al.*, 1991; Meinert *et al.*, 1993; Jochle and Trigg, 1994; Mumford *et al.*, 1995). It has been suggested that GnRH may be more successful than hCG in inducing ovulation in larger, thicker-walled follicles. GnRH also does not have the disadvantage of inducing refractoriness of response due to antibody formation (Mumford *et al.*, 1995). However, much of the work done to date on using GnRH has been in mares during their natural oestrous period, rather than with a synchronized oestrous regime. The limited work done on using GnRH with prostaglandin to time oestrus and ovulation has suggested that there is no significant change in the timing of ovulation as a result of treatment, compared with the use of prostaglandin alone (Voss *et al.*, 1979; Booth *et al.*, 1980; Squires *et al.*, 1981a). Therefore, though the regime suggested in Table 12.7 would be feasible, it is yet to be proved that it is a significant improvement on other regimes.

12.6.2.5. Combination treatments

Several combination treatments are used, some of which have already been mentioned.

The two most commonly used and of current interest, progesterone and prostaglandin and progesterone and oestradiol, are specifically discussed in the following sections.

12.6.2.5.1. PROGESTERONE AND PROSTAGLANDIN. Combination treatments of progesterone and prostaglandin are increasingly popular, as such treatments often improve the timing of ovulation and may reduce the length of progesterone supplementation. Administration of progesterone via intravaginal sponges for 20 days, with a PGF_{2α} injection on the day of sponge removal, was reported to result in oestrus at 1.8–2.2 days and in ovulation at 3.0–5.4 days, respectively, post PGF_{2α} injection (Palmer, 1979; Draincourt and Palmer, 1982). Today, administration of progesterone is normally only for 7–9 days, with prostaglandin administered on the day progesterone treatment ceases. Using this protocol and again with the intravaginal sponges, Palmer (1979) demonstrated that, on average, oestrus occurred earlier (3.8 days) than figures suggested for progesterone treatment alone. The timing of ovulation was, however, very variable, at 8–15 days after prostaglandin injection. Later work by Palmer *et al.*, (1985), using the same sponges but inserted for 7 days, with prostaglandin at sponge withdrawal, suggested better synchrony of ovulation, at 10.1–14.0 days after sponge removal (Table 12.8).

It is evident that yet again considerable variation in response is observed and that the variation may be greater with shorter-term progesterone supplementation during the early breeding season (Hughes and Loy, 1978; Palmer, 1979).

Table 12.7. The timing of oestrus and ovulation in the mare, using two injections of prostaglandin and gonadotrophin-releasing hormone (GnRH) implants. AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0	Prostaglandin
Day 15	Prostaglandin
Day 19	Oestrus commences
Day 21	GnRH implant
Day 22	Ovulation may occur – covering/AI

Table 12.8. The timing of oestrus and ovulation in the mare, using progesterone supplementation, plus prostaglandin. PRID, progesterone-releasing intravaginal device; i.m., intramuscular; AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0–8	Progesterone supplementation (intravaginal sponges or PRID, i.m. injection or oral administration)
Day 8	Prostaglandin
Day 12	Oestrus
Day 16 onwards	Ovulation may occur – covering/AI

12.6.2.5.2. PROGESTERONE AND OESTRADIOL. This combination treatment is increasingly popular. Both hormones may be administered daily via intramuscular injection for 10 days (R. Pryce-Jones and C. McMurchie, Wales, 1997, personal communication) or PRID containing progesterone plus 10 mg oestradiol (held within a gelatin capsule), inserted for 10 days (Rutten *et al.*, 1986). This may be followed, as in the previous protocols, with an injection of PGF_{2α} at the end of the treatment (Table 12.9), and this combination has proved successful, with a reported 81.3% of treated mares ovulating 10–12 days after the PGF_{2α} injection (Loy *et al.*, 1981). Normal pregnancy rates have been reported to AI after such treatment (Jasko *et al.*, 1993). Further refinement of PRID or the development of slow-release subcutaneous capsules may further enhance the use of such combination treatments, removing the need for time-consuming daily injections.

It is evident that the use of oestrous manipulation does not allow precise and

accurate timing of ovulation in the mare. However, there is no evidence to suggest that the use of such treatment significantly affects conception rates (Palmer *et al.*, 1985) and some techniques are more successful than others at allowing the time of ovulation to be predicted. Many of these hormone regimes really only reduce the random spread of ovulations within a population, rather than allowing the exact timing to be predicted. Even so, such an effect allows ovarian examination or teasing to be concentrated into a shorter period of time, so reducing labour costs, etc. Manipulation of the oestrous cycle is therefore regularly practised and, with the additional use of ultrasonography and rectal palpation, can allow an accurate and precise prediction of ovulation to be made.

Many of these synchronization techniques can also be used to induce ovulation and oestrus in mares with some abnormal ovarian conditions. These cases will be discussed further in Chapter 19.

Table 12.9. The timing of oestrus and ovulation in the mare, using progesterone and oestradiol treatment, followed by prostaglandin. AI, artificial insemination. (NB: Considerable variation in the individual mare's response may be observed.)

Time	Drug to be administered/Event
Day 0–10	Progesterone and oestradiol treatment
Day 10	Prostaglandin
Day 20	Ovulation may occur – covering AI

12.6.2.6. Manipulation of breeding activity in the stallion

The breeding season in the stallion, as in the mare, is governed by photoperiod and the stallion reacts to increasing day length in a manner similar to that seen in the mare. The breeding season of the stallion can, therefore, be advanced by the introduction of a 16 h light/8 h dark regime in November/December. Continual stimulation, however, produces refractoriness and a return to normal seasonal changes, despite the altered photoperiod (Argo *et al.*, 1991). Manipulation of stallion reproduction is not as essential as manipulation in the mare, as it is the mare's reproductive cycle that is normally limiting to performance. Given enough encouragement, a stallion will naturally breed during the non-breeding season, but less efficiently.

12.7. Conclusion

It is evident that the preparation of both the mare and the stallion for covering needs considerable thought, especially if the animals concerned are maiden and/or have the added complication of an athletic career behind them. However, providing adequate time and forethought are invested, most horses that are anatomically and physiologically sound are capable of breeding, regardless of their previous careers. The manipulation of reproduction, though widely practised, will not in itself allow the precise timing of oestrus and ovulation. However, if used in conjunction with rectal palpation or ultrasonic scanning, it will allow accurate determination of timing of ovulation and, as such, is a very useful tool in stud management.

13

Mating Management

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13.1. Introduction

Chapters 1–4 discuss in detail the anatomy and physiology of reproduction in the horse and attendant control mechanisms. Reproductive activity is governed by season and commences at puberty, which occurs at between 10 and 36 months of age. The mare will only allow mating to occur when she is in her sexually receptive oestrous phase, which can be referred to as oestrus, season or heat. This period of sexual receptivity occurs regularly at 21 days during the breeding season and lasts 2–10 days (average 5 days). Oestrus is synchronized with ovulation and ensures that the mare is mated or covered at the optimum time for fertilization.

Ovulation occurs normally about 24 h before the end of oestrus, so in an average mare it will be on day 4 of a 5-day oestrus (Ginther, 1992). There is, however, considerable variation between mares. Sperm are reported to survive for 24–72 h within the mare's reproductive tract, but the exact time must remain in doubt, as most experiments on longevity are carried out *in vitro* (Watson and Nikolakopoulos, 1996). Some reports suggest that sperm longevity may be up to 7 days (Newcombe, 1994). The ovum, on the other hand, is thought to survive only 8–12 h, though longer times have been reported in *in vitro* work (Hunter, 1990). The timing of mating is therefore very important, and in practice most mares are covered every 48 h while oestrus lasts (usually twice per oestrus) or until the mare has ovulated.

13.2. Mating Systems

The human desire to control the covering process has led to a wide variety of mating systems, varying from the two extremes of natural covering to intensive in-hand covering.

13.2.1. Natural mating

In the natural system, stallions run with their mares and detect mares in oestrus at will, examining them daily for signs of sexual

receptivity. This process is leisurely and unrushed and the signals used by the stallion to detect oestrus are smell and taste rather than sight (Stone, 1994). Natural courtship may occur over several days, as the mare slowly progresses from dioestrus into full oestrus, and takes place between a mare and stallion that are well known to each other. When she is fully in oestrus and receptive, the courtship culminates in mating. Courtship of a mare in full oestrus follows a sequence of events. The stallion will normally stand and fix his eyes upon a mare that he suspects is receptive, arch his back and neck and draw himself up to his full height, and become restless, pacing around, pawing the ground and stamping his feet. He will show the typical facial grimace, termed flehman, or tasting of the air (olfactory stimulation), often accompanied by roaring or vocalization (Fig. 13.1). He will approach the mare and gauge her response to his attentions. The whole process takes some time, during which the mare, if she is truly receptive, will stand quietly and possibly nicker in response if interested. Once the mare's interest has been ascertained, the stallion will have the confidence to approach her more closely, normally from the head, working his way slowly down her neck, nickering as he does so; he may nudge her slightly or lightly bite her neck. All the time, he is watching for her response and for signs of rejection. If he feels confident, he will then work his way down her flanks and on to her hindquarters and to her perineal area. At any time, he may pause to reassure himself that she is still interested. If all is still amicable, he will nudge her vulva and clitoral area. If the mare is in full oestrus, she will stand still, relatively passive throughout the whole procedure, showing her interest by curling her tail to one side, urinating – often bright yellow urine with a characteristic odour – or she may just take up the urinating stance. She will expose her clitoris by inverting the lips of the labia around the ventral commissure, termed winking (Fig. 13.2) (Ginther, 1992; McDonnell, 1992).

If she is not in oestrus, she will show hostility to the stallion, who will be unable to get



Fig. 13.1. The typical facial grimace, termed flehman, shown by a stallion when in the presence of an oestrous mare.



Fig. 13.2. A mare 'showing' to the stallion by lifting her tail to one side and displaying a passive, quiet reaction to his attentions. Note the inverting of the labia lips around the clitoris, termed winking.

closer to her than the initial advance. In nature, the stallion will then turn away, transferring his attentions elsewhere, and return to her later in that day or on the next (Bristol, 1982; Klingel, 1982; Keiper and Houpt, 1984; Kolter and Zimmermann, 1988).

Once the stallion is sure that the mare is truly receptive, he will mount her. His penis normally becomes erect as he approaches the mare and she shows interest. Mounting will only occur when his penis is fully erect. If the stallion is very sure of himself and the

mare, he will often mount her directly and ejaculate immediately. A stallion with less confidence or one not so sure of the mare may nudge or push her forward slightly prior to mounting or half mount her a couple of times first, in order to ascertain her reaction, before he commits himself and runs the risk of injury. The number of mounts per ejaculate tends to be higher at the beginning and end of the season and in stallions with a low libido (Pickett and Voss, 1972). Ejaculation follows a varying number

of pelvic oscillations, during which time the stallion may dance on his hind feet. Successful ejaculation is signalled by rhythmic flagging of the tail and can be confirmed by feeling for the urethral contractions along the ventral side of the penis. Ejaculation is followed by a terminal inactive phase, when the stallion remains quiescent on the mare while penile erection subsides. The stallion will not naturally dismount until his penile erection and engorgement of the glans penis have completely subsided (detumescence) (Fig. 13.3; see Chapter 2; Boyle, 1992; Davies Morel, 1999).

The time taken to achieve ejaculation varies with stallion and circumstances. On average, a stallion will achieve erection within 2 min of contact with the mare, be ready to mount within 5–10 s of full erection and achieve ejaculation within 5 s of mounting, with a final post-coital quiescent stage of up to 30 s (McDonnell, 2000).

It has been demonstrated, by means of an open-ended artificial vagina, that ejaculation begins as the pelvic thrusts cease and consists of six to nine jets of semen, deposited over 6–8 s, each a result of urethral contraction. The volume of each successive jet decreases, the first three jets making up 70% of the total seminal volume. The remaining jets mainly comprise secretions of the semi-

nal vesicles, the gel fraction of stallion semen (Kosiniak, 1975; Weber and Woods, 1993; Davies Morel, 1999).

In the natural system a stallion will cover a mare in oestrus many times, up to eight to ten matings in 24 h. Nature's system works extremely well, with high pregnancy rates (Bristol, 1982, 1987). It is a system, however, that is rarely practised today – only occasionally seen in pony studs or with horses run on large expanses of land with minimal managerial input. For such a system to run completely naturally, the stallion will only cover the mares within his herd and no outside mares may be introduced solely for covering. The introduction of foreign mares causes a disruption in the hierarchy and can result in jealousy and hostility from other mares and uncertainty between the stallion and the introduced mares (Ginther, 1983c; Ginther *et al.*, 1983). The need to introduce outside mares to a natural mating system is accommodated in some native pony studs by running the stallion out with specific groups of mares. Visiting mares are put in their own group and home-bred mares in another and the stallion is moved around between the groups and allowed to cover the mares at will when they come into oestrus.

Though conception rates are higher in natural breeding than in controlled breeding



Fig. 13.3. The final terminal stage after ejaculation, where the stallion remains inactive as detumescence occurs (Derwen International Welsh Cob Stud).

(Bristol, 1987; Davies Morel and Gunnarsson, 2000) the natural breeding system has many disadvantages from the breeder's point of view, when maximum financial return from the stallion is required. The natural system limits the number of mares the stallion can cover in a season, as each mare gets covered eight to ten times per oestrus, whereas, if mating is timed correctly, in theory only a single service is required. As a result, breeders feel the need to control events to protect the investment made, maximize the number of mares covered per season and minimize the risk of injury to stock. This interference is at the root of many of the problems associated with stud management today.

13.2.2. Mating in hand

Mating in hand is practised by the majority of studs today and involves complete control over the events surrounding covering. In general, we now control the life of the horse to such an extent that there is very little similarity between the natural life and the one imposed.

It is common practice to segregate fillies and colts early on in their lives, sometimes from birth, often at weaning. Naturally, colts and fillies would run together as part of a herd. Colts would be disciplined by other mares and stallions, socially interact with fillies and learn respect at a young age, when the chance of serious damage is reduced (Bristol, 1982). In the intensive systems of today, this social introduction of the young stallion with fillies and mares is removed. In addition, we expect stallions to cover mares that are unknown to them. The failure to develop social awareness and respect for mares results in stallions being inept at the interaction prior to covering (McDonnell, 2000). When this is coupled with the strangeness of any unknown mare and the sexual tension present, it is not surprising that the risk of mare rejection and injury to both parties is high. An additional consideration is the significant value of some stallions and the need to control events so as to protect one's investment – the stallion – and to mini-

mize the number of coverings per pregnancy so as to optimize his use and maximize financial return.

To overcome the potential danger to stallions and to optimize their use, most systems mate in hand. Humans therefore have total control over the number of mares covered by each stallion (Umphenour *et al.*, 1993). However, one of the major drawbacks of this system is that there is the need to interpret the mare's oestrous signals before the stallion is allowed near her. As discussed previously, the stallion uses the senses of smell and taste to detect mares in oestrus; the breeder, however, has to rely on the sense of sight only (Palmer, 1979; Stone, 1994). This method leads to inaccuracies and can prove unreliable. Hence a teaser stallion is employed.

13.3. Teasing

Teasing is the use of a stallion, often not the one chosen for mating, to encourage a mare to demonstrate oestrous behaviour under controlled conditions. The principle being that, as soon as the mare is thought, by visual signs, to be in oestrus, she is brought in contact with an entire male horse under controlled conditions in an attempt to enhance the signs of oestrus and confirm the initial diagnosis. Once the mare is confirmed as being in true oestrus, she can then be prepared for covering by the chosen stallion. This system allows the stallion destined to cover the mare to have access to her only when she is in true oestrus and at the most optimum time for fertilization, so minimizing the risk of injury and maximizing the chance of conception (Squires, 1993b). Apart from allowing oestrus to be detected, teasing is now known to play a role in enhancing reproductive activity. Prolonged teasing results in elevated gonadotrophin-releasing hormone concentration, so increasing gonadotrophin release in both the mare and the stallion, thus advancing ovulation and encouraging uterine clearance in the mare and enhancing libido in the stallion (Irvine and Alexander, 1991; Lieberman and Bowman, 1994; McDonnell and Murray, 1995).

The teaser stallion is often kept purely to detect mares in oestrus. He is often of low value. If he is injured or damaged by an objecting mare, then there is no significant loss. Often a pony stallion is used. In many native-pony-breeding studs, the stallion to be used for covering also acts as the teaser for his mares. This allows the reproductive state of the mare to be confirmed, but does not give the stallion the protection of using a separate teaser. However, such stallions tend to be less valuable, native mares show oestrus more readily and the mare may well be teased initially over a teasing board or equivalent, providing some protection.

There are problems associated with in-hand covering, mainly because the courtship that naturally takes place over a prolonged period is concentrated into a short space of time and forces the attentions of the stallion upon the mare. Some mares object to such forced attentions, even if they are in full oestrus, and such objection may mask the signs of oestrus. Mares with foals at foot often object, occasionally violently, to the removal of the foal prior to teasing and covering, a practice normally carried out to protect the foal. In the natural system, the foal would still be in the close vicinity of the mare, but seems to know instinctively to keep its distance. Teasing some mares before feeding or turnout can also give erroneous results, and environmental conditions of extreme heat, cold, rain or wind may mask signs of oestrus. Some mares need a longer time of teasing before they can be coaxed into demonstrating oestrus and, in a busy stud working to a tight schedule, there is little time for extended teasing and hence she may never seem to be ready to cover. Again, some mares will only show oestrus under certain circumstances, i.e. only in the covering yard, when the perineum is being washed, when the tail is bandaged prior to service or when a twitch is applied. This is where the mare's records are invaluable in identifying any such idiosyncrasy (Lieberman and Bowman, 1994). There is also evidence that mares may have a preference for certain stallions and this may be associated with vocalization. It has been suggested that the

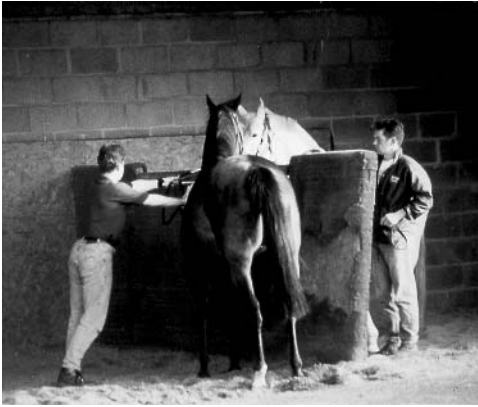
more vocal a stallion, the greater his popularity (Pickerel *et al.*, 1993).

When a mare is in oestrus she will be docile, accept the attentions of the stallion, take up the urination stance and expose her clitoris (referred to as 'showing') and demonstrate a general lack of hostility towards and signs of acceptance of the stallion (Figs 13.2 and 13.4a–d). There are a wide range of methods used to tease mares, depending on the stud, the value of the stock and the facilities available.

13.3.1. Trying board

One of the most common methods of teasing is a trying or teasing board. The mare and stallion are introduced one on either side of the board and their reactions monitored. The board is designed so as to provide protection for both and should be high enough to allow just the horse's heads and necks to reach over. It is solid in construction, often made of wood and ideally twice the length of the horses. Its top should be covered by curved rubber or equivalent, to provide protection if the stallion or mare attempt to attack each other over the board (Fig. 13.4a–d).

The approach of the teaser to the mare over the board should mimic that of the natural approach. Initially muzzle to muzzle, the teaser is then allowed to stretch his muzzle along the mare's neck, possibly gently nipping her. The attitude of the mare to this attention is closely observed; signs of hostility include laid-back ears, squealing, biting and kicking out, indicating that the mare is still in dioestrus. In contrast, leaning towards the stallion, raising of the tail and the other typical signs of oestrus indicate that she is ready to be covered. If the mare is interested, then her flank can be turned towards the trying board and the teaser allowed to work his way further down her body. It is, however, very important that direct contact with the mare's genital area is avoided, in order to prevent possible disease transfer to other mares teased. After a few minutes of such attention, most mares will show definite signs of oestrus; some mares, as discussed, may take longer.



(a)



(b)



(c)



(d)

Fig. 13.4. A mare being teased over a trying or teasing board. The mare and stallion should be introduced initially muzzle to muzzle and the stallion allowed to work his way down to the mare's vulva (a, b, c). If she is in oestrus, she will show little, if any, objection. A mare not in oestrus will usually object violently (d). (Photographs Elizabeth Wood.)

Using the same principle, a stable door may act as an alternative to a purpose-built trying board, with the teaser in the stable and the mare introduced to him outside the door. This is a popular practice in the smaller, more native-type studs, but can be dangerous unless the teaser is well known and is unlikely to be overzealous (Fig. 13.5).

13.3.2. Teasing over the paddock rail

Teasing over the paddock rail provides an alternative popular system, used in larger studs. The teaser is led in hand to the pad-

dock rail of fields containing mares, which are running loose, normally in small groups. A permanent trying board is often built into the paddock rail or a movable trying board is placed there. The reaction of the mares to the teaser is noted. Most mares in oestrus will approach the teaser by the fence and show definite signs of oestrus, others may show hostility and some may appear disinterested. Mares that show no reaction, often due to shyness or low social ranking, can be caught and brought up to the trying board to be tested individually. Those that show interest can also be tried individually for confirmation or brought in for covering immediately.



Fig. 13.5. Providing the stallion is of a known good temper, then he may tease the mares he is to cover over his stable door.

This is an efficient method for use with mares that are turned out, as it greatly reduces time and labour. It is best to avoid teasing mares by this method immediately after turnout or just before feeding, as this can give erroneous results.

Walking the teaser past the paddock fence on a regular basis, a version of the above, is of particular use if the teaser can be ridden or requires regular exercise in hand. His daily route can then be organized to pass appropriate paddocks and the general reaction of the mares observed. Those that show interest are then caught and taken to the covering yard.

This system does have potential problems when mares have foals at foot. Conflicting reports state either a considerable danger to the foal or that foals distance themselves, as they do naturally, and that mares, especially in small groups, are very careful to avoid damage to their foals.

Not all mares react to these two forms of teasing. Those that are less demonstrative may well be missed, especially if they are low within the hierarchy (Lofstedt, 1988). It is essential in such systems, therefore, that a detailed record of the mare's normal oestrous behaviour is available.

13.3.3. Teasing in chutes and crates

In the southern hemisphere, including South Africa and South America, mares tend to run in large herds and are handled less frequently. In such enterprises, the mares may be run into chutes or crates and held individually for a short period of time and teased from outside. This system reduces labour and enables large numbers of mares to be teased in as a short a period of time as possible, with limited handling. The mares need to be accustomed to this system or errors may result (Fig. 13.6).

13.3.4. Teasing pen

A further alternative system is to confine the teaser in a railed or boarded area in the corner of a paddock. This system is particularly popular in Australia and South America. The area confining the teaser normally has high boards with a grill or meshed fencing, possibly with a hole through which the teaser can put his head (Fig. 13.7). An alternative is the use of a small pony or miniature-breed stallion confined by a stout fence. If he escapes, it is not too disastrous, as his size limits his ability to cover the mares, though it is not impossible. These are again good systems for teasing a large number of mares, but they do require frequent observation of mares for signs of oestrus and, as with teasing over the paddock rail, it may be difficult to pick up shy mares. It must, of course, be remembered that the teaser can only be confined in the railed area for relatively short periods of time.

Along the same lines as the teasing pen is a central teasing pen surrounded by a series of individual pens in which mares can be held.



Fig. 13.6. Teasing of mares using a chute in the USA. Mares may be placed in the crate individually or the crate incorporated as part of a chute system. The stallion is restrained outside the crate and the mares stopped momentarily next to the stallion to gauge their response (photograph Julie Baumber).



Fig. 13.7. Use of a teasing cage confines the stallion or teaser and allows any mares, either loose within the field or restrained, to approach at will (photograph Julie Baumber).

A further variation on this theme is the use of two adjacent boxes, divided by a grill, one for the stallion and another for the mare. Yet another involves the teaser being confined in a stable in the corner of a yard with the mare free within the yard to show to the stallion at will. Such an arrangement is used at the National Stud, Newmarket, UK (Fig. 13.8).

These last three systems are good for use with difficult mares, as they can be left alone

to show in their own time without competition from other mares and can be observed from a discrete distance.

13.3.5. Vasectomized stallions

Rarely, vasectomized stallions may be used to run out with mares. This can be especially useful with maiden or difficult mares; those



Fig. 13.8. An arrangement such as this can be used to tease shy or reluctant mares. The stable in the corner of a covering yard (open window in background) is used to confine the teaser, while the yard (in the foreground) allows the mare to be free to exhibit oestrus at leisure (The National Stud, Newmarket).

mounted can then be covered by the intended entire stallion. This system has the obvious danger of running unaccustomed mares and stallion out together and is therefore of limited use especially with valuable stock. However, it is an extremely reliable method of detecting oestrus and has the reported advantage of increasing the number of mares showing regular 21-day cycles (Barnisco and Potes, 1987). The other major disadvantage is that this system allows intromission to occur, and so enables the transfer of venereal disease. This can be averted by surgical retroversion of the penis, causing it to extend caudally (between the hind legs) at erection, making intromission impossible (Belonje, 1965). Venereal-disease transmission via the stallion's muzzle is not, however, eliminated.

13.3.6. Hermaphrodite horses and androgenized mares

Hermaphrodite horses are very useful as teasers but are very rare and therefore not really a viable alternative. Androgenized mares (mares treated with testosterone) have been used successfully, but are not common practice (McDonnell *et al.*, 1986).

13.3.7. Teasing mares with a foal at foot

A mare with a foal at foot may present problems. The foal often becomes agitated with the unaccustomed attention to its mother and so distracts her. To overcome this, the foal may be penned, held within reach or sight of its mother or removed completely from sight and sound while teasing occurs (Fig. 13.20). Prior knowledge of a mare's normal behaviour in such circumstances is very useful.

13.3.8. Conclusions on teasing

In any system of teasing, direct contact between the stallion's muzzle or penis and the mare's genitalia must ideally be avoided. Direct contact risks the transfer of disease to successive mares via the stallion. This is one of the major advantages of teasing over a trying board and a potential disadvantage of many of the other methods discussed.

Not all mares show under the above systems and some require specific management, prolonged individual teasing etc. The key to success is careful observation, as it must be remembered that all mares react individually and no system is 100% reliable. Therefore, further confirmation of a mare's reproduc-

tive activity is often required. This is achieved by veterinary examination.

13.4. Veterinary Examination

Routine veterinary examination to confirm a mare's reproductive state is used in many studs, especially those running valuable stallions with the aim of optimizing their use. Veterinary techniques may be used alone or to back up teasing and confirm diagnosis and optimize the timing of covering. There are three types of veterinary examination that may be used in this context: ultrasonic scanning; rectal palpation; and vaginal examination. All three techniques have been previously described (Chapter 11). They are used to assess ovarian, uterine, cervical and vaginal activity. This assessment can be used to confirm a mare's sexual state, correlate coitus to ovulation and diagnose venereal infections. For all techniques, the mare should be restrained in stocks, with the perineal area thoroughly washed and her tail bandaged up out of the way (Fig. 13.9).

13.4.1. Ovarian assessment

The main activity assessed in the context of covering management is ovarian activity. The techniques used are ultrasonic scanning and rectal palpation (Chapter 11). In particular, the presence of follicles and/or corpora lutea (CL) and their consistency, appearance and position are noted so that the time of ovulation can be estimated and the most appropriate time for covering determined.

Follicles may develop on either ovary and often several follicles develop, some of which regress due to an inability to react to hormonal stimulation (Chapter 3). As oestrus and ovulation approach, normally one to three follicles can be identified as dominant. The diameter of these follicles used to be advocated to be a good predictor of the imminence of ovulation (Greenhof and Kenney, 1975). However, though diameter may be used as a guide, preovulatory follicular diameter varies considerably. Most mares

ovulate follicles of 3.5–4.5 cm in diameter, others habitually ovulate follicles of 6.0–6.5 cm and other mares as small as 2.0 cm (Ginther and Pierson, 1984a,b; Pierson and Ginther, 1985b; J. Newcombe, Wales, 2001, personal communication). Multiple ovulations are seen; double ovulations have been reported to occur in 23% of ovulations in the thoroughbred (Davies Morel and O'Sullivan, 2001) but are rarer in other breeds, such as draught and Arab mares, and are very uncommon in native ponies (Arthur and Allen, 1972; Newcombe, 1995). If multiple ovulations are present, they are likely to ovulate at a smaller size. The presence of multiple ovulation may well preclude a mare from covering. Despite the increasing success of managing twin conceptuses by pinching out (see Chapter 14), many studs still prefer not to cover multiple-ovulating mares but rather to leave them and advance the



Fig. 13.9. Stocks for use in restraining mares for internal veterinary examination (Monarch Manufacturing, King Street, Willenhall, West Midlands, UK).

next oestrus, which will hopefully demonstrate only a single ovulation (see Chapter 12). Of further note with regard to multiple ovulations is that they may occur asynchronously but both may still be fertile. As such, scanning or rectal palpation may indicate a single dominant follicle but scanning several days later may indicate multiple CL and a multiple pregnancy.

Ovulation occurs in two stages (see Chapter 1). These two stages can be used as an additional guide to the imminence of ovulation (J. Newcombe, Wales, 2001, personal communication). As ovulation becomes increasingly imminent, the follicular wall becomes thinner and the pressure of the follicular fluid contents decreases. On rectal palpation, this decline in follicular pressure can be felt. On scanning, the clear spherical shape of the follicle becomes less clear and the margins become thicker and more 'ragged' in appearance (Ginther and Pierson, 1984a,b; Pierson and Ginther, 1985b; Sertich, 1998). Such follicles would be expected to ovulate within 24 h and most studies advise that the mare should be covered immediately. The best pregnancy rates are achieved by covering within the 48 h period prior to ovulation (Woods *et al.*, 1990;

Katila *et al.*, 1996). If at re-examination the mare has not ovulated, she should be re-covered immediately. In such a scenario, human chorionic gonadotrophin is often administered to advance ovulation (Chapter 12). The prescence of a CL within the ovary indicates that the mare has ovulated and that, unless the ovulation is very recent, within 12 h, there is little point in covering the mare. Ova only remain viable for up to 12 h after ovulation (Ginther, 1992), whereas sperm may remain viable for up to 7 days (Newcombe, 1994). Coverings later than 12 h after ovulation result in higher rates of early embryonic death, with pregnancy rates for covering more than 24 h after ovulation at zero (Woods *et al.*, 1990; J. Newcombe, Wales, 2001, personal communication). CL appear as semi-solid structures within the cavity of the old follicle. At scanning, they are evident as grey spherical shapes (Figs 13.10 and 13.11; Ginther and Pierson, 1984a,b; Pierson and Ginther, 1985a).

At rectal palpation, a new CL can be detected as a soft friable mass. At 24 h after ovulation, they feel firmer and a pit may be felt. Later on, when assessing via rectal palpation, they may be hard to distinguish from follicles.

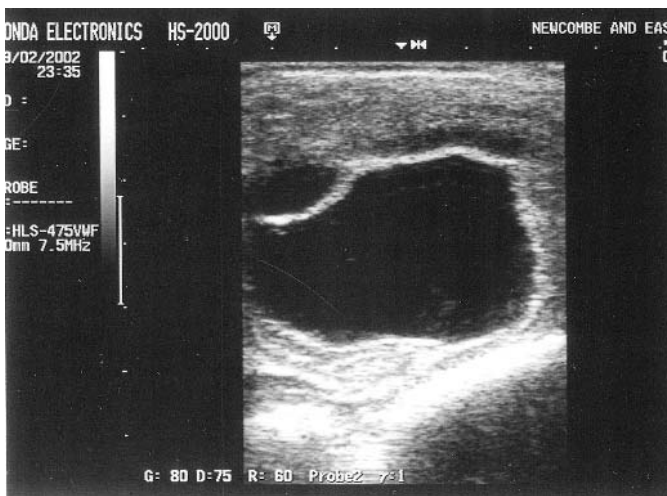


Fig. 13.10. An ultrasonic scanning photograph of a follicle immediately prior to ovulation; notice the thick wall and the loss of a clear spherical shape (photograph Dr John Newcombe).



Fig. 13.11. An ultrasonic scanning photograph of a corpus luteum immediately after ovulation (photograph Dr John Newcombe).

13.4.2. Uterine assessment

Uterine activity and appearance can be used as a further guide to reproductive activity. As with ovarian activity, this is assessed via scanning and rectal palpation. Striking changes occur within the uterus between oestrus and dioestrus. This is thought to be due to the presence or absence of progesterone, rather than oestrogens. Once the dominance of progesterone has been removed, as the mare goes into oestrus, the uterine endometrial folds become oedematous and can be clearly visualized at scanning (Ginther and Pierson, 1984a,b; Hayes *et al.*, 1985; Ginther, 1992; Plata-Madrid *et al.*, 1994; Bergfelt, 2000). The dense central portions of the folds appear echogenic (white/grey) and the oedematous portion non-echogenic (grey/black). Hence, when the uterine horn is viewed in cross-section, it resembles a sliced orange or cartwheel (J. Pycock, Wales, 2000, personal communication). This endometrial oedema can be scored and, as such, bears a close correlation to behaviour scores (Hayes *et al.*, 1985; Squires *et al.*, 1988). However, more recent work, by Plata-Madrid *et al.* (1994), indicates that oedema may peak up to 6 days prior to ovulation. Despite this, uterine oedema is used in practice as a useful indicator of imminent

ovulation and is a good indicator of basal progesterone levels and hence the likelihood of elevated oestrogens and oestrus.

Rectal palpation may also be used to indicate uterine changes associated with reproductive activity. The tone, size and thickness of the uterus are assessed, tending to increase under the dominance of progesterone; the uterus during oestrus appears smaller and more flaccid (Ginther, 1992; Bergfelt, 2000).

13.4.3. Cervical and vaginal examination

Cervical and vaginal examination is a less reliable determinant of reproductive activity; nevertheless, it can be a useful tool in confirming a mare's sexual state. The cervix and vagina are visualized via a vaginoscope (Chapter 11). The cervix during oestrus appears pink to glistening red in colour, is relaxed and appears to 'flower' into the vagina, with its lining oedematous. Shortly after oestrus, the cervix begins to contract and becomes paler pink in colour, with a thick secretion. By dioestrus, the cervix is closed, pale in colour and dry. Pregnancy may be considered an extreme form of dioestrus; as such, the cervix is tightly closed

and white to pink in colour, with a central, sticky, mucous plug. Vaginal secretions originate from, and so mimic, cervical secretion. During oestrus, the vagina appears red/pink in colour with fluid secretions; during dioestrus secretions appear sticky and viscous, causing the vaginal walls to adhere together and thus making speculum examination more difficult. The vaginal secretion during pregnancy is thick, thickening even more as pregnancy progresses.

13.4.4. Hormone profiles

It has been suggested that declining oestradiol plasma concentrations can be used as an indicator of imminent ovulation (Allen *et al.*, 1995). However, due to individual variation, this is unlikely to be consistently diagnostic in itself but may, like cervical and vaginal examination, be an additional aid in diagnosis.

13.5. Preparation for Covering

Once it has been determined that the mare is in oestrus and ready for covering and that she has the appropriate negative swab certificates (see Chapter 11; Horse Race Betting Levy Board, 2001), attention must be turned to the preparation of the mare and the stallion for covering. Preparation depends entirely upon the system used for mating and varies from the strict codes of practice within the Thoroughbred industry to practically no preparation at all in the case of many native-pony studs. The most cautionary preparation will be considered in the following account. Other studs dispense with some, if not all, of the preparation techniques. Exact management also depends on the size of the stud, labour available, etc. In larger studs, there are often two to three breeding sessions per day, spaced at regular intervals, e.g. 9.00 a.m., 2.00 p.m. and 7.00 p.m. Most mares are covered twice within an oestrus, at 24–48 h intervals or until ovulation has been confirmed (Ginther, 1992; Newcombe, 1994).

13.5.1. The mare

The mare is prepared with all eventualities in mind. She is bridled and restrained, her tail bandaged and the perineal area washed thoroughly. When washing the mare, gloves should be used and there should be a different swab of cotton wool for each swipe. Each swipe should be taken from the buttocks towards the perineum and the cotton wool discarded immediately to prevent contamination of the washing solution. If soap is used it should be mild, non-detergent soap and the area should be thoroughly rinsed. Soap and disinfectants can act as spermicides and may also upset the natural microflora of the genital tract, opening up the opportunity for colonization by opportunistic bacteria and hence endometritis (see Chapter 19). A hose is a good alternative to manual washing and is popular in the USA.

At this stage, the vulva of any mares with Caslick vulvoplasty should be opened (episiotomy). If any sutures remain, these should also be removed to avoid damage to the stallion. Once the mare has been washed, she is led to the covering area, where felt covering boots may be fitted to her back feet, which should have had their shoes removed prior to arrival at the stud (Fig. 13.12).

She may also have a nose or ear twitch applied, depending on her temperament and past behaviour. Some studs twitch as standard, believing that prevention is better than cure as far as damage to expensive stallions is concerned. If she has the reputation of being particularly bad-tempered, she may need one of her forelegs held up in a carpal flexion, or hind-leg hobbles may be fitted to prevent her lunging forward and objecting when the stallion mounts. Hobbles are particularly popular in the USA and also in Australia, but they are not commonly used in the UK. It is questionable whether a mare requiring such drastic restraint is truly in oestrus. In such cases, conception rates may be adversely affected, due to both the incorrect timing of service and the stress of such treatment. Other articles used occasionally include blinkers, hood or blindfold, especially for highly-strung maiden or difficult

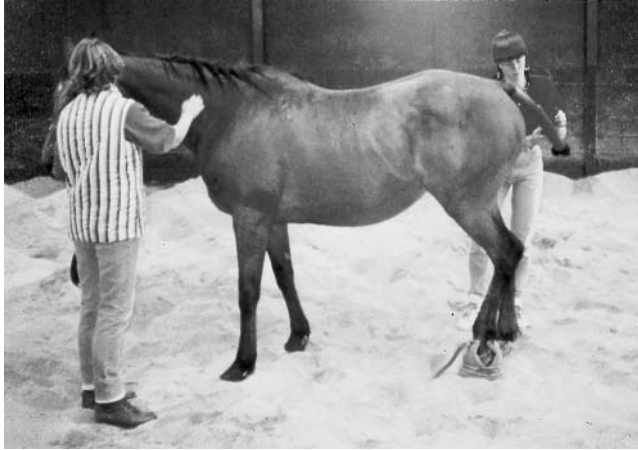


Fig. 13.12. Mare ready for covering, tail bandaged, perineum washed and wearing covering boots (photograph Angela Stanfield).

mares. A mare being covered by a stallion that tends to bite his mares can be protected by use of a shoulder or wither pad, with or without a biting roll (Fig. 13.13).

On very rare occasions, tranquillizers may be administered 15–30 min prior to covering; this can be of use in particularly nervous or vicious mares. If such extreme restraint is necessary, then the use of such a mare for breeding should be questioned, as there is a risk that such a temperament is heritable.

13.5.2. The stallion

A stallion's behaviour during the covering season can be unpredictable and therefore dangerous. Management techniques can reduce this danger, such as by the regular tying up (racking up) of stallions in their stables for a period of time each day. This makes catching an enthusiastic stallion much safer when a mare arrives on the yard, if he has been tied up and is used to the routine. The use of a specific bridle for exercise and



Fig. 13.13. Additional items used in covering, from the left: a twitch, separate biting roll, breeding roll, withers pad, covering boots and neck guard with biting roll (National Stud, Newmarket).

for covering is advised, so that a stallion is aware of what is required of him by the tack presented. For covering in intensive in-hand systems, a stallion is normally tacked up with his covering bridle and long rein; alternatively, a pole may be used giving a greater degree of control.

He is then led from his box to a washing-down area, where his penis, genital area, belly and inside hind legs are washed with warm clean water. Ideally, the penis should be erect at washing. Erection may occur naturally, due to the anticipation of covering; others may require initial teasing. Antiseptic and/or soap solutions, once very popular, should be used with care, due to their spermicidal effects (Betsch *et al.*, 1991; Fig. 13.14). The stallion is then led to join the mare waiting in the covering area.

13.5.3. The mare and stallion

This preparation of the mare and stallion, as described, represents the extreme of full precaution to avoid all possible transfer of bacterial infection and is referred to as the minimal-contamination technique (Kenney *et al.*, 1975a). In the Thoroughbred industry, where the value of stock is high, such precautions are economically justifiable. In other

studs, with progressively lower turnovers and less valuable stock, preparation becomes less cautionary in nature. It is interesting to note that several of these procedures are gradually being introduced into the less intensive studs. For example, many breed societies now require swabbing prior to covering and some restrict the use of the yearly premium stallions to mares with negative swab certificates.

The other extreme to that practised in the Thoroughbred industry is seen in many native studs, where no preparation of the mare or stallion is practised, except the possible bridling of the pair for restraint and removal of the mare's hind shoes. Such extensive systems run a high risk of disease transfer. However, one of the saving graces is that native-type stock appear to suffer less from genital infections, though of course they are not completely immune.

13.6. Covering

The style and management of the actual covering procedure varies considerably. In-hand covering is the norm within the Thoroughbred industry in most countries and, with the increasing value of stock, in-hand breeding is now widely practised.

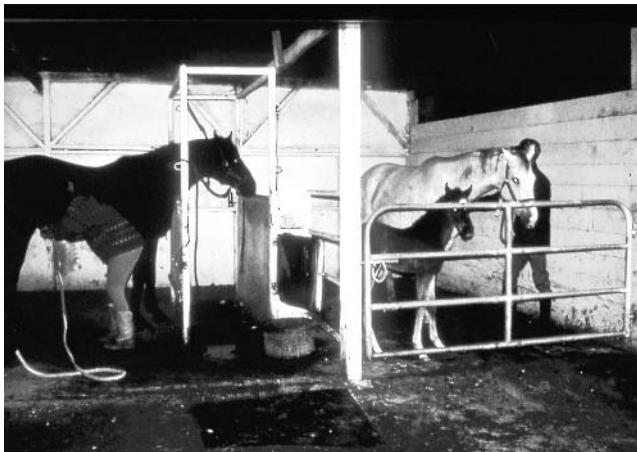


Fig. 13.14. Prior to covering, a stallion's penis and genital area should be washed; erection can be encouraged by the close proximity of an oestrous mare (photograph Julie Baumber).

For each horse, there should be at least one handler, ideally wearing stout footwear and a hard hat. The stallion handler should also carry a stick for reprimanding and, in many traditional systems, there is also an assistant to help the stallion gain intromission, if necessary, or to hold the mare's tail. A fourth handler may also be present to hold the mare steady and prevent her tottering too far forward. It is essential that all handlers know their exact role and that an emergency procedure is drawn up beforehand. Once prepared, the mare is normally taken into the covering area first to await the arrival of the stallion. The stallion is then brought in to cover the mare immediately or, in some systems, the stallion may initially be introduced to the mare over a trying board positioned within the covering area. This provides him with protection during initial contact and is required by some stallions to avoid a prolonged period with the mare in the covering area before full erection and mounting occurs, a potentially dangerous situation. The mare is then led away from the trying board ready for covering. At this stage, she may be further restrained by means of a twitch or hobbles. Once the mare is ready, the stallion is allowed to approach her, normally at an angle several feet from her nearside to avoid startling her and causing her to kick out.

Once the stallion's penis is fully erect, he should be allowed to mount. Mounting before full erection should not be allowed. There are two blood pressures within the stallion's penis at erection: turgid and intromission pressure (Chapter 2). Attainment of intromission pressure is essential to avoid damage to the stallion on entry into the mare (Fig. 13.15).

As the stallion is allowed to mount the mare, all handlers should stand to one side of the animals, usually the left. If problems do occur, then both the stallion and the mare can be pulled towards their handlers, turning their hindquarters away from anyone who is likely to be kicked.

As the stallion mounts the mare, she will probably totter forwards. This is to be allowed as long as she does not move too far.

Excessive tottering will put a strain on the stallion, who will rest more of his weight on her back and also cause her possible strain or injury. Tottering too far forward can be prevented by placing the mare up against a protective barrier. This arrangement may also be used to protect handlers with mares that strike out (Fig. 13.15f). If a leg strap is being used to restrain the mare, it must be released at this stage. For this reason, tight hobbles are not advocated, as they do not allow the mare to move in order to accommodate the weight of the mounting stallion.

Many traditional studs still advocate the use of an assistant stallion handler to pull the mare's tail to one side and guide the penis into the vagina. However, this is no longer considered good practice, as any unexpected external stimuli may disrupt the normal sequence of events leading up to ejaculation.

Successful ejaculation is signalled by the rhythmic flagging of the stallion's tail (Fig. 13.15e). In most stallions this is clearly evident, but, if in doubt, the urethral contractions can be felt by a hand placed along the ventral side of the penis. However, possible disruption of the natural sequence of events must again be considered.

After ejaculation, the stallion should be allowed to relax on the mare (Figs 13.3. and 13.15g). During this period of relaxation, the glans penis, which has increased in size considerably at ejaculation, returns to normal, allowing the penis to be withdrawn. Early withdrawal can cause damage to the stallion and mare. He should then be allowed to dismount at will, which may take several minutes. The mare should be turned towards her handler and walked slowly away, allowing the stallion to slide off and reducing the chance of him being hit should she lash out. The stallion should similarly be turned towards his handler after he has dismounted, again reducing the chance of injury to the mare or handlers. Some studs routinely collect a dismount semen sample from the drips on the penis at dismount. The presence of spermatozoa confirms that ejaculation has taken place. At this time a dismount swab may also be taken from the urethral area. Increasingly, in studs running high-value stallions, the mat-



(a)



(b)



(c)



(d)



(e)



(f)

Fig. 13.15. The sequence of events associated with in-hand covering: (a) the stallion is bridled and brought into the covering area to meet the mare; (b) the mare may be teased by the stallion over a trying board immediately prior to covering; (c) covering boots are put on the mare's hind feet; (d) the stallion is allowed to cover the mare – ideally, the stallion should be calm and not overenthusiastic; (e) ejaculation is marked by flagging of the stallion's tail; (f) the mare may be stood up against a trying board during covering to prevent excessive tottering forward; (g) (*opposite*) ejaculation is followed by a quiescent period prior to dismount (The Elms Stud; photographs Victoria Kingston).



(g)

Fig. 13.15. *Continued.*

ing process is videoed to provide additional evidence that covering has taken place.

After covering, the stallion should be allowed to wind down and walked to the washing area. There his penis should be washed and his genitalia examined for abrasions. Many practitioners advocate walking the mare around slowly after covering to prevent her straining and so losing semen. However, as the vast majority of the semen is deposited into the top of the cervix and into the uterus, beyond the cervical seal, it is very unlikely that there will be significant loss at post-coital straining. Any semen that is lost is likely to have been that deposited into the vagina and, as such, will have limited bearing on fertility.

Occasionally, the mare and stallion differ in size. If so, certain precautions should be taken. A very large stallion put on to a small mare should be considered carefully, as it may cause her problems in late pregnancy, due to large fetal size near term. The reverse situation poses no such problems. In order to assist in the mating of unequal-size horses, some covering yards have a dip in the floor or use a breeding platform to equalize the horse's heights (Fig. 13.16). In the USA hydraulic breeding platforms are becoming popular and can be adjusted to the exact height required.

A breeding roll may also be used if there is concern that the stallion's penis is too

long or with maiden mares. The breeding roll is placed between the mare's perineum and the belly of the stallion, preventing him from thrusting too far and so causing damage. The breeding roll is usually a padded leather roll of 15–20 cm in diameter and of the order of 50 cm long. In an attempt to maintain sterile conditions, it can be covered with a disposable examination glove (Fig. 13.13).

13.6.1. The covering yard

The covering yard can be any area that is quiet, dry and safe with a non-slip surface and away from the mêlée of the general yard. A paddock, open yard or specially designed covered area (Fig. 13.17) may be used. If large enough, this covering yard can double up as an exercise area (Fig. 13.18).

If an area is to be specially designed, it should be at least 20 m by 12 m, roofed, with two sets of wide doors to allow horses to enter and exit in different directions. It is very important that the floor of the covering yard is clean, non-slip and dust-free. Suitable surfaces include clay, chalk, peatmoss, woodchip or, nowadays, rubber matting, which is becoming popular (Fig. 13.19). The requirement for a non-slip surface makes floorings such as concrete inadvisable, though they are occasionally used.

Fig. 13.16. A horseshoe-shaped breeding platform may be used to increase the height of a small stallion, so easing the covering of a tall mare.



Fig. 13.17. Covering in a field or paddock is common practice in smaller studs (photograph Gillian Humphries).

13.6.2. Management of foal-heat covering

A perceived major problem when covering mares is the danger to any foals at foot (Fig. 13.20). The majority of mares foal at the stud or arrive with foals of a very young age. In nature, this presents no significant problems, as foals naturally move away from the mat-

ing activity but stay within the vicinity of the mare. If teasing and/or mating is to occur in a large open area, the foal may therefore be let loose and will normally keep well away from the proceedings, but within sight of its dam, reducing anxiety and stress. If mating is to occur in a small enclosed covering yard or if the mare is difficult, the foal should be



Fig. 13.18. The covering yard, if large enough, can double up as an exercise area (Derwen International Welsh Cob Stud).

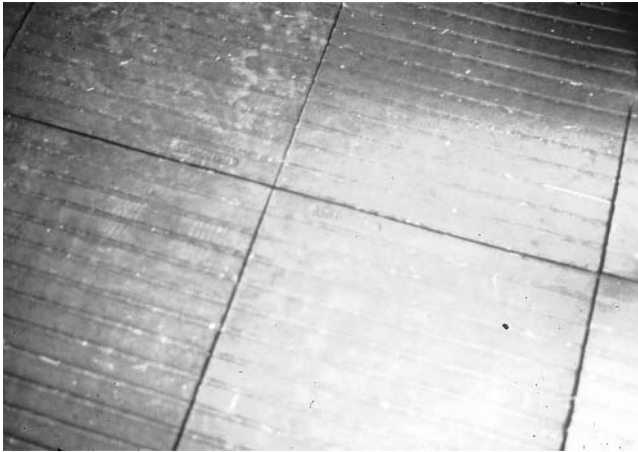


Fig. 13.19. Rubber matting may also be used as a suitable, clean, dust-free, non-slip surface in a covering yard.

restrained for its own safety. It may be removed and put in a loose box within sight of the mare and with a handler to watch it. However, some mares will become distracted by the antics of the foal. In such cases, more success may be achieved by removing the foal altogether, out of sight and earshot. The system used depends entirely on the character of the mare and foal, the facilities available and personal preference. Further consideration of foal-heat breeding is given in Chapters 16 and 19.

13.6.3. Management of mares susceptible to post-coital endometritis

All mares suffer from post-coital endometritis to some extent, but some mares appear to be more susceptible than others and, in such a situation, conception rates can be significantly depressed. These mares can be managed in a number of ways in order to reduce the post-coital inflammatory response; details of these treatments are given in Chapter 19.

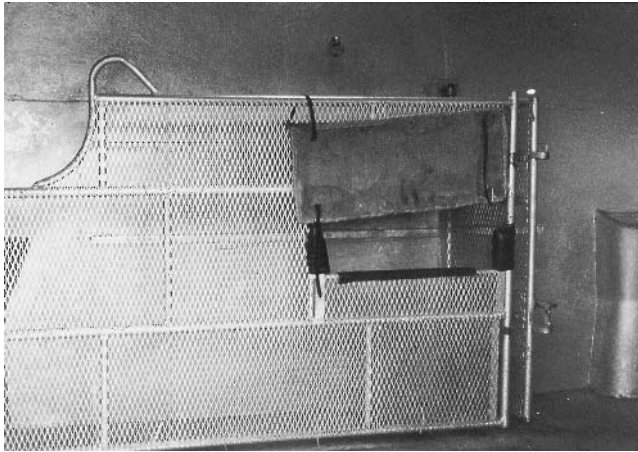


Fig. 13.20. Mares with foals at foot may prove difficult to tease and cover successfully. In this case, the cage can be used to hold the foal, allowing reasonable access by the dam but ensuring the foal's safety during teasing and covering (photograph Julie Baumber).

13.7. Variations in Mating Management

As mentioned previously, there are many variations on the above theme, many being more cost-effective and used on the smaller native studs. The alternatives include covering in a small open paddock, yard or railed area with a non-slip surface, possibly earth or grass. Many stallions, especially in the Welsh-cob industry, successfully cover mares in hand in a convenient field near to the main yard. This is how mares would have been covered historically, when stallions were walked around from farm to farm. If covering is to occur in a large field, then the stallion used must be well behaved and controlled, as his escape may incur considerable wasted time and disturbance to mares in adjacent fields.

The other extreme to in-hand breeding is pasture breeding, where the stallion is allowed to run free with his mares to cover them at will (Bristol, 1987). This system is the nearest to the natural situation but, as discussed at the beginning of this chapter, presents problems with visiting mares. Pasture breeding is popular in South Africa and South America, where mares are run in large herds over wide expanses of land, with stal-

lions running out freely (Bristol, 1982; Ginther, 1983c; Ginther *et al.*, 1983).

A halfway house between in-hand breeding and pasture breeding is to allow the stallion to be loose in the field and introduce the mares to him individually, either restrained or free. This system allows visiting mares to be successfully covered in a system that is quite near to the natural one. Stallions used in this system must be well behaved, of an appropriate temperament and their characteristics well known by the mare handler, so that any necessary avoiding action may be taken.

13.8. Conclusion

It is evident that there is a vast array of management practices for covering mares. The system ultimately chosen is up to the individual stud, its facilities, normal practices and labour available. Financial considerations also have a considerable bearing on management choices. Intensive breeding systems are increasingly employed, providing more protection for both handlers and horses, but often at the expense of natural equine behaviour and reproductive performance.

14

Management of the Pregnant Mare

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14.1. Introduction

Before the management of the pregnant mare is considered, it is essential to ascertain whether she is indeed pregnant. Once pregnancy has been confirmed, the mare can be monitored and managed accordingly. The diagnosis of a twin pregnancy poses specific problems in the mare and these will also be considered.

14.2. Pregnancy Detection

Pregnancy detection may be carried out for a number of reasons. Ideally, it is conducted as soon as possible so that non-pregnant mares can be re-covered on their next oestrus and twin pregnancies identified and appropriate action taken. It may also be necessary for sale or for insurance purposes, particularly relevant if the mare is in the last two-thirds of her gestation. Finally, if the agreement at covering is no foal, no fee, it establishes whether a fee is due. The customary date on which the buyer of a stallion nomination has to pay in the northern hemisphere is 1 October; the corresponding date in the southern hemisphere is 1 April.

The mnemonic AEIOU describes the major aspects of an ideal pregnancy test:

- A Accurate
- E Early
- I Inexpensive
- O Once only
- U Uncomplicated

There are also problems with many tests in that some fail to indicate embryo viability or

the presence of twin pregnancies, to anticipate fetal death or to differentiate between embryonic vesicles and uterine cysts. Numerous methods of detection are available; however, none as yet meet all the above criteria (Collins and Buckley, 1993; Lofstedt and Newcombe, 1997; Henderson *et al.*, 1998).

14.2.1. Manual

Manual methods of pregnancy detection are the oldest and cheapest and still in common use. A major advantage is that they give immediate results, though they can only be accurately used after day 20 *post coitum*. Examination is carried out via rectal palpation or cervical/vaginal examination.

14.2.1.1. Rectal palpation

Rectal palpation, as described in Chapter 11, allows the uterus to be felt through the rectum wall. Initial work on this technique was carried out by Day (1940). This and subsequent work suggests that, with experience, an accurate diagnosis can be made 20–30 days *post coitum*. Detection of pregnancy is reported to be possible at day 16, but poor accuracy precludes its use so early. Occasionally, with awkward mares, diagnosis is not possible until day 50. Normally, the accuracy of detection is very good and at its best at day 60, at which stage the age of the fetus can be estimated to within 1 week and the presence of twins detected (Van Niekerk, 1965; Bain, 1967; Roberts, 1986c). Table 14.1 indicates the size of the embryonic vesicle at various stages throughout early pregnancy.

Table 14.1. The size of the embryonic vesicle at various stages during early pregnancy in the mare.

Age of pregnancy (days <i>post coitum</i>)	Size of embryonic vesicle (diameter in mm)	Comments
15	15–20	Embryonic vesicle first detected
20	30–40	
30	40–50	
40	60–70	
50	80–90	
60	100–130	Most accurate diagnosis

Between days 16 and 20, a thickening of the uterine wall, rather than identification of the embryonic vesicle, may indicate pregnancy. Between day 20 and day 60, the embryonic vesicle can be felt as a discrete swelling either side of the midline, at the junction of the uterine horn and uterine body. Additionally, the uterus may feel turgid, rather than flaccid as in the non-pregnant state.

After day 60, the fetus appears to move towards the uterine body and, due to its size, palpation of the margins of the fetal sac is difficult. In the case of twins, the fetal sacs merge and their diagnosis becomes less accurate. From this stage onwards, the whole uterus becomes progressively less turgid and more distended, with no discrete swellings

detected (Fig. 14.1). Hence the accuracy of pregnancy detection at these later stages declines. However, as pregnancy progresses still further, it becomes possible to feel fetal structures, such as the head and ribs, through the uterine wall.

After day 200, accuracy of detection returns to near 100%. At this stage, the fetus can be readily felt through the now much thinner uterine wall (McKinnon, 1993). Pregnancy may also be obvious from the mare's external appearance.

Pregnancy detection using rectal palpation is most commonly conducted between days 20 and 35 of gestation and is now considered accurate and safe, though some historical discussion with regard to a link with abortion has occurred (see section 14.2.1.3).

Fig. 14.1. The expected size of the fetal sac at various stages of pregnancy when undertaking rectal palpation. From the top left to bottom right: non-pregnant uterus, 25-, 45- and 60-day pregnant uterus.

The major disadvantage of rectal palpation is that it cannot be accurately used early enough to detect mares failing to conceive in time to allow them to be re-covered within one oestrous cycle. However, the technique is quick, simple and cheap and gives immediate results.

14.2.1.2. Cervical/vaginal examination

Vaginoscopy – the examination of the mare's cervix and vagina via a speculum (Chapter 11) – can be used as an aid to pregnancy detection, but is not accurate enough to be used in isolation (Roberts, 1986c). The pregnant cervix is white/light pink in colour, firm and with a sticky mucus. The vagina also contains sticky opaque mucus, making the insertion of the speculum difficult. Most cervical and vaginal changes are evident from day 17–20 onwards. However, there is significant variation in cervical/vaginal characteristics between mares, making the technique inaccurate when used in isolation (Asbury, 1991). Used in conjunction with rectal palpation, however, it can be a useful aid to diagnosis in awkward mares.

14.2.1.3. Abortion risk

There are conflicting reports associating manual manipulation of the mare's reproductive tract with an increased risk of abortion. Allen (1974) and Voss and Pickett (1975) produced no evidence of such an association in pony mares. In fact, they suggest that abortion of a twin by squeezing (pinching out) of the fetal sac through the uterine wall *per rectum* at day 40 requires considerably more relative force than ordinary rectal palpation, but is reported to carry only a minimal risk to the remaining fetus. However, Osborne (1975) demonstrated that uterine myometrial activity does increase at palpation and that increased levels of such stress are associated with abortion. The current consensus of opinion is that any association between rectal palpation and abortion is due to the stress of unaccustomed handling, rather than the technique *per se*.

14.2.2. Blood tests

Blood tests are of use in small ponies and mares with injury or rectal/vagina tears, which render rectal palpation and ultrasonic pregnancy detection not feasible. Blood plasma concentrations of one or several hormones that may be used to indicate pregnancy. These tests can be very accurate but have the disadvantage of high costs and a possible delay before the results are available. The tests used may be biological, immunological or chemical, relying mainly upon the detection of equine chorionic gonadotrophin (eCG), progesterones and oestrogens.

14.2.2.1. Equine chorionic gonadotrophin

As discussed in Chapter 3, between days 35 and 120 of gestation, the endometrial cups produce eCG (pregnant mare serum gonadotrophin), which can be detected in plasma samples between days 40 and 100 *post coitum* (Allen, 1969a,b). The presence of eCG was traditionally detected by biological tests, relying upon its effect on the reproductive tract of laboratory animals, such as the mouse (McCaughey *et al.*, 1973; Asbury, 1991).

In the 1960s a new immunological or antibody test was developed, called the haemagglutination inhibition test, now more commonly known as the mare immunological pregnancy test (Wide and Wide, 1963; Allen, 1969a). This, and the more recent immunological tests, such as ELISA and radioimmunoassay (RIA), are the tests now widely used. During the period days 45–100 *post coitum*, all three tests have an accuracy of 60–100%. The ELISA test, however, allows the earliest detection, at day 35, with 43% accuracy and is more accurate (100%) at day 40 than the other two tests (31–37% accuracy) (Squires *et al.*, 1983b). All three tests can be performed in less than 2 h.

Despite its accuracy, testing for eCG has the major disadvantage of being unable to determine whether the fetus is viable. After spontaneous or induced abortion, the endometrial cups continue to secrete eCG and therefore an erroneous positive result may be obtained (Mitchell and Betteridge, 1973; McKinnon, 1993).

14.2.2.2. Progesterone

Progesterone is the hormone responsible for the maintenance of pregnancy and, as might be expected, elevated levels are indicative of the presence of a fetus. When testing very early in pregnancy at the time of the mare's possible return to oestrus, elevated levels (those greater than 3–4 ng ml⁻¹) on day 16–17 *post coitum* are 71% accurate in indicating pregnancy. In non-pregnant mares, progesterone levels should be declining at this time. However, prolonged dioestrus can be mistaken for pregnancy (Holtan *et al.*, 1975a,b; Hunt *et al.*, 1978; Villani *et al.*, 2000). Commercial kits are now available for the rapid testing of plasma progesterone concentrations.

14.2.2.3. Oestrogens

Oestrone sulphate is one of the major oestrogens identified in the mare's plasma during pregnancy (Cox and Galina, 1970; Raeside *et al.*, 1979). Elevated levels are observed at day 35–60; however, at this stage, they are of limited diagnostic value but may be a preliminary indicator of fetal viability (Hyland and Langsford, 1990). However, elevated plasma oestrone sulphate concentrations from day 85 onwards, peaking around day 210 (Terqui and Palmer, 1979), provide an accurate, but late, diagnosis of a viable pregnancy. Shuler (1998) reported that plasma concentrations higher than 1.6 ng ml⁻¹ are diagnostic of pregnancy and concentrations less than 0.8 ng ml⁻¹ confirm a negative result. Concentrations between these two levels are inconclusive. Biological tests for oestrogens have largely been replaced by the chemical (Cuboni) test and now the immunological RIA and ELISA (Cuboni, 1934; Terqui and Palmer, 1979; Evans *et al.*, 1984). Oestrone sulphate analysis will also indicate fetal viability, as the fetal-placental unit is required for its production (Lasley *et al.*, 1990; Stabenfeldt *et al.*, 1991). Despite this, the method is of no use for early diagnosis.

14.2.2.4. Early pregnancy factor

An early pregnancy factor (EPF) has been identified, using a rosette inhibition test in

several animals as early as 6 h *post coitum* (Shaw and Morton, 1980). An equine EPF has been identified from 2 days after ovulation. After embryo transfer or embryonic death, EPF declines to non-pregnancy levels within 2 days. As such, this test has the potential to provide a useful tool not only for detecting pregnancy, but also for monitoring *in vivo* viability of equine embryos and for detecting embryonic death (Gidley-Baird and O'Neill, 1982; Takagi *et al.*, 1998). Early detection of pregnancy failure would allow the mare to be prepared for covering on her next natural oestrus or at an artificially accelerated return to oestrus. As yet, the test is not commercially available. If diagnosis of fertilization ever becomes routinely possible as early as 6 h *post coitum*, this will allow the mare to be re-covered at that same oestrus, giving her a second chance to conceive.

14.2.3. Urine tests

Urine tests are not very popular but do have their uses in non-lactating mares where rectal palpation or blood sampling proves difficult (Cox, 1971; Evans *et al.*, 1984).

14.2.3.1. Oestrogens

Oestrogens in the mare, being of a relatively small molecular weight, are able to pass unaltered through the kidney's filtration system and can therefore be detected in the urine. Other hormones, such as eCG, are too large and therefore cannot be detected in urine. As with plasma oestrogen concentrations, pregnancy detection is possible from about day 90, although accuracy of diagnosis at this stage is low. By day 150, accuracy improves significantly (greater than 95%) (Boyd, 1979).

14.2.4. Milk tests

Plasma hormone concentrations are most commonly used, but there may be occasions in lactating mares when obtaining a milk sample is easier and less stressful.

14.2.4.1. Progesterone

Progesterone is the major hormone that can be isolated in milk for use as a pregnancy test. Milk progesterone concentrations increase in parallel with plasma concentrations. Elevated levels are observed after oestrus; a subsequent decline after 5–10 days is indicative of no pregnancy, but continued elevated levels indicate pregnancy.

14.2.4.2. Oestrogens

Conjugated oestrogens can also be identified in milk, the concentration pattern again correlating closely to that observed in plasma and urine samples (Sist, 1987; Raeside *et al.*, 1991).

14.2.5. Faeces test

Unconjugated oestrogens have been isolated in the faeces of pregnant mares from day 120 and so may be used as a late pregnancy test. Such a test may be useful for feral mares (Bamberg *et al.*, 1984; Sist, 1987).

14.2.6. Ultrasonic pregnancy detection

In recent years, ultrasonic techniques have revolutionized the detection of pregnancy in many animals. Ultrasonic detectors are based on the principle that ultrasonic sound waves are absorbed or reflected by the objects they hit. The relative amount of absorption or reflection depends on the density and movement of the object and can be transduced into a visual image. This method has the advantage of giving an immediate result and of usually being done on the stud, as the equipment is fully portable. More detailed accounts of the general reproductive use of ultrasound are given in Chapters 11, 12 and 19 and in reviews by other authors (Chevalier and Palmer, 1982; Allen and Goddard, 1984; Ginther, 1984; McKinnon, 1993; Sertich, 1998).

14.2.6.1. Doppler system

Fraser *et al.* (1973) initially developed the use of Doppler ultrasound. This machine enables movement, and hence fetal heartbeat, to be detected, along with uterine arterial blood flow. The ultrasonic signal is emitted from, and received by, a transducer placed on the mare's abdomen or within her rectum via a rectal probe, and is transduced into an audible sound. The fetal heartbeat can be heard from day 42 of gestation onwards as a distinct beat, accelerated in comparison with that of the mare, though it is not consistently heard until day 120. The enhanced blood flow through the pregnant mare's uterine artery can also be heard at the same time. It is present as a distinct whooshing noise at a slower beat than the fetal heart. This characteristic blood flow through the pregnant uterus is diagnostic in itself.

This method is very accurate at day 120, but accuracy in early pregnancy is not guaranteed and thus it cannot be successfully used within the timescale required for the mare to be returned to the stallion at her next oestrus. The time span of use is similar to that of the test for eCG, but it has the obvious advantage of detecting a viable foal (Fraser *et al.*, 1973; Mitchell, 1973).

14.2.6.2. Visual echography (ultrasonic pregnancy detection)

Hackeloer (1977) developed the use of visual ultrasonic echography. The reflected ultrasonic waves in this system are transduced into a visual picture on a visual display unit. Examination is *per rectum* by means of a probe carrying the ultrasonic emitter and transducer (Fig. 14.2). The uterus is examined and within it the embryonic vesicle (Roberts, 1986a; McKinnon and Carnevale, 1993; McKinnon *et al.*, 1993). The embryonic vesicle can be detected from day 11–12 onwards as a discrete spherical sac (Ginther, 1984). This typical spherical nature of the equine trophoblast and its characteristic position, after day 17, at the junction of the uterine horn and body is fortunate and, as in the case of the human, allows detection at an early stage



Fig. 14.2. The transducer probe of the ultrasonic scanner is placed in the rectum of the mare and angled down towards the uterus. The image produced is displayed on a visual display unit.

with an accuracy in excess of 98% (Ginther, 1989a; Hallowell, 1989). This method can also be used in the accurate detection of twins in early pregnancy, in addition to uterine cysts, fluid accumulation, sacculations, etc., or, at a later date, fetal viability.

The image produced by ultrasonic echography illustrates hard structures, such as bone, which reflect sound waves, as white, and illustrates fluid, which absorbs sound waves, as black, with variations in between.

The size of the embryonic vesicle at various stages of early pregnancy is given in Table 14.1. Pregnancy can first be detected at day 11–12, at which stage only the embryonic vesicle can be identified. At this stage, the embryo is mobile and migrates within the uterus, making detection more difficult. In addition, there is a higher risk of embryo mortality in day 11 embryos than in older ones. By day 17–18, the embryo has become 'fixed', normally at the junction of the uterine body and uterine horn, and so identification is easier (Ginther, 1983a,b; J. Newcombe, Wales, 2001, personal communication). By day 20, the embryo itself can be identified within the embryonic vesicle (Fig. 14.3). Day 24 heralds the first detection of the fetal heartbeat (Allen and Goddard, 1984; Ginther, 1984) and between days 59 and 78 fetal sex may be first determined (Curran and Ginther, 1989).

In practice, most studs scan for pregnancy at day 18, which is the most accurate time and early enough to allow arrangements to be made for re-covering. An additional scan at day 40, after the period of highest risk, may be considered. In the Thoroughbred industry, where there is a high incidence of twins, initial scanning is normally carried out at day 11–12, in order to identify twins and manage the mare accordingly (section 14.3). The mare is then rescanned again at day 18–20 and again at day 40.

14.2.7. Fetal electrocardiography

A fetal heart electrocardiogram may be obtained from electrodes strategically placed on the mare's body, which pick up the fetal electrical heart impulses. The read-out given can be used to detect the presence of a fetal heartbeat and also any abnormalities (Colles *et al.*, 1978; Rossdale and Ricketts, 1980; Roberts, 1986a). Its popularity as a tool for diagnosing pregnancy is limited, due to the complication of setting it up, and it can only be used in late pregnancy. However, it does have its uses in detecting and monitoring fetal stress or cardiac abnormalities, especially near to parturition or during a difficult delivery (Schott, 1993). It can also confirm fetal viability and the presence of twins (Parkes and Colles, 1977; Buss *et al.*, 1980).

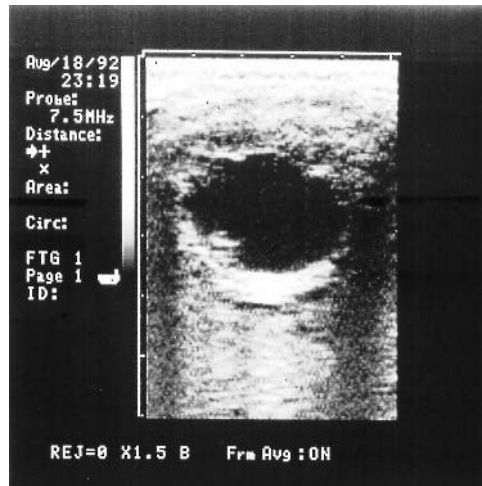


Fig. 14.3. A typical scanning photograph showing a 20-day-old pregnancy. The embryo is evident as a white structure towards the bottom left of the dark fluid-filled area.

14.3. Management of Twin Pregnancies

The conception of twins is a significant and increasing problem in pregnant mare management. As discussed previously (Chapter 5), the mare is monocoelous, the uterus being unable to adequately support two pregnancies. Twin pregnancies rarely survive to term, most commonly resulting in abortion in mid- to late pregnancy (9–10 months) (Rossdale, 1987; Ball, 1993b). They are the most common cause of non-infectious abortion (Roberts and Myhre, 1983), accounting for 20–30% of all occurrences (Torbeck, 1986). Only 9% of twin pregnancies survive to term; 64.5% of these cases result in two dead foals, 21% in one live foal and 14.5% in two live foals (Card, 2000). Rates of twinning differ significantly between breeds but, in the Thoroughbred, twin rates have been reported to be as high as 16.2% (Newcombe, 1995). As such, it is advantageous to identify and manage twin pregnancies early on, especially if re-covering is planned. The advent of ultrasonic scanning has significantly helped the early identification of twins.

There are four main management practices used to reduce the incidence of twins: monitor ovulation; wait and see; manually reduce; or treat with prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$) (Card, 2000). Historically, the incidence of twinning was reduced by monitoring ovarian activity, using rectal palpation, and withholding covering from mares with more than one large follicle. The mare would then be covered on the next natural or the next artificially advanced oestrus. This successfully reduced twinning rates within a population, but with it conception rates declined and the time interval between parturition and successful covering increased (Miller and Woods, 1988; Pugh and Schumacher, 1990). In order for these drawbacks to be addressed, identification and treatment of actual twin pregnancies, rather than potential twin pregnancies, are required. Naturally, in excess of 83% of twin pregnancies are spontaneously reduced to singles around the time of embryonic fixation (day 18) (Ginther, 1987; Ginther and Bergfelt, 1988). So one option is to monitor the pregnancy and observe if natural reduction occurs. If not induced, abortion at a later

stage may be advocated. The advent of scanning now allows such monitoring to occur easily. An alternative to natural reduction is to manually reduce. Manual reduction of twins to a single has been reported to be up to 96% successful between days 13 and 16 (Pascoe *et al.*, 1987a,b; Ginther, 1989a,b). Manual reduction involves the manual squeezing of the smallest embryo, identified by ultrasound, either between the thumb and forefinger or by using the scanner probe to push the conceptus against the uterine wall and pelvis until the vesicle ruptures (Ginther, 1987; Pascoe *et al.*, 1987a; Rossdale, 1987). This is best done prior to fixation (day 18) and so is normally carried out at initial scanning on day 11–12 (Ginther, 1989b). After fixation, the manual reduction of bilateral twins (one in each horn) can still be very successful, but reduction of unilateral twins (both in the same horn) runs a higher risk of losing the whole pregnancy (Ginther, 1989a, b). Other methods of manual reduction at a later stage of pregnancy (after day 40) have been reported, such as ultrasound-guided allantocentesis and transabdominal fetal cardiac puncture, but have not proved as successful as early manual reduction (Card, 2000). An alternative to manual reduction of one twin is to artificially induce abortion of the whole pregnancy and re-cover the mare at the next advanced oestrus. Abortion and subsequent return to oestrus and ovulation can be induced using a single injection of PGF_{2α}; multiple injections may be required later in the pregnancy. Abortion can be induced prior to the next expected oestrus, i.e. before day 21 of pregnancy. This is often done at the time of first scanning, which may be as early as day 11, in which case the mare's return to pregnancy after abortion may be delayed by only 15 days or so. Alternatively, the pregnancy may be allowed to progress longer in the hope that natural reduction may occur before PGF_{2α} is required. However, if a rapid return to oestrus is required, then PGF_{2α} must be administered prior to the development of the endometrial cups at day 40.

14.4. Embryonic Loss

Mares suffer from relatively high rates of early embryonic death, particularly in the first 40 days of pregnancy. The reasons for and causes of embryonic loss and the subsequent failure to produce a foal are discussed in detail in Chapter 19. In an attempt to reduce embryonic loss, some studs routinely place their mares on daily progesterone supplementation therapy in the form of injections or oral progesterone treatment for the first 100 days of pregnancy. From day 100 onwards, placental progesterone should be adequate. The use of such routine progesterone supplementation is questioned by some, as there is little evidence that it prevents embryonic mortality in the majority of pregnancies and it prevents the expression of the warning signs of a return to oestrus should embryo mortality occur (Darenius *et al.*, 1987; Holtan, 1993).

14.5. Pregnant-mare Management

The general management of the pregnant mare is important in order to ensure that the fetus is given every advantage and that the mare's future reproductive capacity is maintained. A fit condition throughout pregnancy is advantageous allowing a mare to foal with ease and be in optimum physical condition to embark on another pregnancy as soon as possible post-partum. An understanding of the developmental changes of both the mare and the fetus during gestation is essential in order to gear her management towards optimum reproductive performance. These developmental changes are detailed in Chapter 5.

There is little specific information on the environmental factors that affect fetal well-being. However, good management of the pregnant mare is clearly one, and can be reviewed under six main headings: exercise; nutrition; parasite control; vaccination; teeth care; and feet care.

14.5.1. Exercise

Exercise and nutrition are closely related and together are the major determining factors of body condition. As with most things, it is the extremes that can prove harmful, and so in the case of exercise a happy medium is to be aimed for (Rossdale and Ricketts, 1980). The absolute level of exercise depends on the individual mare and her history. A moderate exercise regime may be provided in the form of self-exercise by regular turnout in an open field or gentle ridden exercise in early pregnancy. Mares that are used to being ridden can be ridden with increasing gentleness up until 6 months of pregnancy. In the last trimester of pregnancy, forced exercise should cease and ideally be replaced by group pasturing. Mares turned out in groups tend to exercise more effectively than those turned out alone (Kiley-Worthington and Wood-Gush, 1987).

Gentle exercise promotes and enhances the circulatory system. As discussed in Chapter 5, the fetus depends entirely upon its dam for its nutrient intake and waste output. This transport system is provided by the blood circulatory system, in particular the utero-ovarian artery and vein. Blood circulation through this system is enhanced by exercise, hence increasing oxygen and nutrient

supply to the fetus. Exercise reduces water retention, or oedema, often associated with mares that are kept inside for prolonged periods of time, especially in late pregnancy. Exercise helps to maintain body condition and reduce obesity; hence the chance of complications at parturition is reduced and the maintenance of muscle tone will enhance delivery (Huff *et al.*, 1985; Fig. 14.4).

Exercise must be consistent and not exhaustive, as this has been associated with high abortion rates. Excessive exercise may also cause stress, as may sudden movements, travelling, sale rings and even low-flying aircraft. Stress is known to increase abortion rates, especially in early pregnancy – around day 40 – as well as during the last 6–8 weeks of pregnancy.

14.5.2. Nutrition

The nutritional requirements of a pregnant mare vary according to whether she has a foal at foot and is lactating, or whether she was previously barren and so has only to satisfy the requirements for her own maintenance plus fetal growth (Hintz, 1993a). The length of lactation in the mare depends today on our interference, but normally runs for 6–8 months. It is, therefore, during this time that



Fig. 14.4. Exercise is essential in order to maintain a mare's fitness – body condition score of 3 – and to prevent circulatory problems in the later stages of pregnancy (Penpontbren Welsh Cob Stud).

the two groups of mares with or without foals at foot vary in their requirements. In an ideal system, and providing there are enough mares to justify it, these two classes of mares should be kept and fed separately. During pregnancy, a mare's body condition and weight should be carefully monitored to ensure that neither excess weight is gained, nor does she have to mobilize her own body reserves to supplement inadequate nutrition. Her feeding management should vary accordingly. The use of a weigh pad is ideal to monitor weight on a regular basis, though a weigh band can still give an accurate indication of weight change. Assessment of body condition by eye and feel can also be undertaken (Fig. 14.5), a condition score of 3 should be maintained throughout pregnancy.

The following discussion concentrates on the nutritional management of the non-lactating mare, as the management of the lactating mare is addressed more fully in Chapter 16. The same general principles of nutrition apply to feeding the mare as to any formulation of a horse ration. The important nutrients are protein, energy, vitamins and minerals, and they should all be balanced according to need. During pregnancy, requirements do not significantly increase until the last 3 months, when they should be

adequate to allow a 14.5% increase in mare body weight. During early pregnancy, the mare's weight gain should be minimal, though, as she progresses into the last trimester (last 100 days), she should be expected to gain in the order of 0.25 kg day^{-1} (Frape, 1998; Card, 2000). This level of weight gain ensures that it is due to an increase in fetal weight and not to excessive deposition of internal body fat. Overfat condition in mares during late pregnancy causes excessive pressure on the internal organs at parturition, as well as limiting uterine size and so reducing fetal birth weight and possibly postnatal viability. Limited fat deposition in late pregnancy is, however, desirable to act as a temporary store for emergency mobilization during early lactation. Failure to gain the required weight may mean that the mare's limited fat reserves of pregnancy are already being mobilized, reducing the energy available to the fetus and hence its growth *in utero*. However, it has been reported that mares restricted to up to 55% of National Research Council requirements (see Tables 14.3–14.5) give birth to normal birth-weight foals. This demonstrates that healthy mares possess the ability to compensate for low nutritional intake without significant detriment to the foal (Frape, 1998).



Fig. 14.5. Though a weigh pad is the ideal, a weigh band can be used as a quick and convenient method of monitoring a mare's weight, especially when used in conjunction with assessment of body condition by eye and feel.

As in the case of other animals, feed must be of good quality, with adequate roughage to aid digestion and prevent the development of vices through boredom (Winskill *et al.*, 1996). Any good stud will, as a matter of routine, have all batches of hay, haylage, etc. analysed to ascertain the dry matter (DM) content, protein, energy, vitamins and mineral concentrations. This allows accurate balancing of feeds available and identifies the appropriate supplements needed. The quality of feed is also important; conditions such as fungal contamination can cause abortion (Pugh and Schumacher, 1990). General poor nutrition has been associated with prolonged gestation, developmental abnormalities and decreased birth weights. These problems are exacerbated if low nutrition levels are evident in late pregnancy (Comline *et al.*, 1975; Silver *et al.*, 1979). Excess body weight has

been associated with uterine inertia and dystocia (Varner, 1983), but this is disputed by others (Henneke *et al.*, 1984).

14.5.2.1. Protein

Levels of crude protein (CP) of the order of 656 g CP day⁻¹ (10.0% (100% DM)) are required in early pregnancy (National Research Council, 1989; Frape, 1998). However, as pregnancy progresses into the last 90 days protein intake needs to increase in parallel with requirements. Levels of protein of the order of 866 g CP day⁻¹ (10.6% (100% DM)) are required in the last 3 months of pregnancy (Tables 14.2–14.5). This level may be achieved by feeding good fresh grasses, but dried grass or hay tends to lose protein in the drying process. Legume hay – for example, lucerne – tends to have a higher

Table 14.2. The average composition of feeds commonly used in horse diets. DM, dry matter; DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium; Na, sodium; S, Sulphur. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Feed	DM (%)	DE (Mcal kg ⁻¹)	Crude protein (%)	Lysine (%)	Fibre (%)	Ca (%)	P (%)	Mg (%)	K (%)	Na (%)	S (%)	Vitamin A (iu kg ⁻¹)
Lucerne	91.0	2.07	17.0	–	25.5	1.24	0.22	0.32	1.42	0.11	0.26	41,900
(hay)	100.0	2.28	18.7	–	28.0	1.37	0.24	0.35	1.56	0.12	0.28	46,000
Barley	88.6	3.26	11.7	0.40	4.9	0.05	0.34	0.13	0.44	0.03	0.15	817
(grain)	100.0	3.68	13.2	0.45	5.6	0.05	0.38	0.15	0.50	0.03	0.17	922
Sugar beet	91.0	2.33	8.9	0.54	18.2	0.62	0.09	0.26	0.20	0.18	0.20	88
(pulp)	100.0	2.56	9.8	0.60	20.0	0.68	0.10	0.28	0.22	0.20	0.22	97
Clover	88.4	1.96	13.2	–	27.1	1.22	0.22	0.34	1.60	0.16	0.15	9,727
(hay)	100.0	2.22	15.0	–	30.7	1.38	0.24	0.38	1.81	0.18	0.16	11,005
Fish-meal	91.7	2.93	62.2	4.74	0.7	5.01	2.87	0.15	0.71	0.41	0.53	–
	100.0	3.20	67.9	5.17	0.8	5.46	3.14	0.16	0.77	0.44	0.58	–
Oats	89.2	2.85	11.8	0.39	10.7	0.08	0.43	0.14	0.40	0.05	0.21	44
(grain)	100.0	3.20	13.3	0.44	12.0	0.09	0.38	0.16	0.45	0.06	0.23	49
Pea	89.1	3.07	23.4	1.65	5.6	0.21	0.41	0.12	0.95	0.22	–	285
(seeds)	100.0	3.45	26.3	1.86	6.3	0.14	0.46	0.14	1.06	0.25	–	320
Sorghum	90.1	3.21	11.5	0.26	2.6	0.04	0.32	0.15	0.37	0.01	0.03	468
(grain)	100.0	3.56	12.7	0.29	2.8	0.04	0.36	0.17	0.41	0.01	0.15	520
Soybean	89.1	3.14	44.5	2.87	6.2	0.35	0.63	0.27	1.98	0.03	0.41	–
(extracted meal)	100.0	3.53	49.9	3.22	7.0	0.40	0.71	0.31	2.22	0.04	0.46	–
Timothy	88.9	1.77	8.9	–	30.0	0.43	0.20	0.12	1.61	0.01	0.12	18,964
(hay)	100.0	1.99	9.7	–	33.8	0.48	0.23	0.13	1.82	0.01	0.13	21,340
Wheat	89.0	2.94	15.4	0.56	10.0	0.13	1.13	0.56	1.22	0.05	0.21	1,048
(bran)	100.0	3.30	17.4	0.63	11.3	0.14	1.27	0.63	1.37	0.06	0.24	1,177

Table 14.3. The daily nutrient requirements of pregnant mares of varying weights. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Pregnant mares	Weight (kg)	DE (Mcal)	Crude protein (g)	Lysine (g)	Ca (g)	P (g)	Mg (g)	K (g)	Vitamin A (10 ³ iu)
9 months	200	8.2	361	13	16	12	3.9	13.1	12
	500	18.2	801	28	35	26	8.7	29.1	30
	700	23.6	1039	36	45	33	11.3	37.8	42
	900	26.8	1179	41	51	38	12.9	42.9	54
10 months	200	8.4	368	13	16	12	4.0	13.4	12
	500	18.5	815	29	35	26	8.9	29.7	30
	700	24.0	1058	37	46	34	11.5	38.5	42
	900	27.3	1200	42	52	38	13.1	43.6	54
11 months	200	8.9	391	14	17	13	4.3	14.2	12
	500	19.7	866	30	37	28	9.4	31.5	30
	700	25.5	1124	39	49	35	12.3	40.9	42
	900	29.0	1275	45	55	41	13.9	46.3	54

Table 14.4. Nutrient concentrations in total diets for pregnant mares (100% dry matter (DM)). Values assume a concentrate feed containing 3.3 Mcal kg⁻¹ and hay containing 2.0 Mcal kg⁻¹ DM. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Pregnant mares	DE (Mcal kg ⁻¹)	Diet proportions		Crude protein (%)	Lysine (%)	Ca (%)	P (%)	Mg (%)	K (%)	Vitamin A (iu kg ⁻¹)
		Conc. (%)	Hay (%)							
9 months	2.25	20	80	10.0	0.35	0.43	0.32	0.10	0.35	3710
10 months	2.25	20	80	10.0	0.35	0.43	0.32	0.10	0.36	3650
11 months	2.40	30	70	10.6	0.37	0.45	0.34	0.11	0.38	3650

Table 14.5. The expected feed consumption of pregnant mares (% body weight).

Pregnant mares	Forage	Concentrate	Total
Maintenance	1.5–2.0	0–0.5	1.5–2.0
Mares in late gestation	1.0–1.5	0.5–1.0	1.5–2.0

protein, even after drying, and so may be adequate for the late pregnant mare.

Protein can be supplemented by the addition of animal products or plant products. Appropriate animal by-products include fish-meal, bone-meal, etc. Such products tend to be expensive and have now been banned in some countries, in the light of the bovine spongiform encephalitis scare in cattle. The other alternatives are plant products, such as

soybean meal, linseed meal, etc. These tend to be cheaper and more popular.

The total protein content of a diet is not the only important factor in satisfying the protein requirements; protein quality is also important. The component parts of all proteins are amino acids, some of which are essential and others of which can be synthesized by the body from other amino acids. The latter are termed non-essential amino

acids. Certain protein-rich supplements may be lacking in specific essential amino acids, so that, even though the total protein content is high, its use to the body is limited. Barley, oats and linseed are all high in total protein content but are lacking in one or more essential amino acids. Soybean meal, on the other hand, contains all the essential amino acids required for the development of the fetus and is therefore a very useful protein supplement for use in late pregnant mares.

14.5.2.2. Energy

Energy requirements increase significantly in late pregnancy but are also important during early pregnancy, their deficiency being implicated as a cause of embryo mortality (Potter *et al.*, 1987). Again, the energy demand of mares in good condition in early pregnancy may be met by good-quality forage. Levels of $16.4 \text{ Mcal day}^{-1}$ ($2.25 \text{ Mcal kg}^{-1}$ (100% DM)) are required (National Research Council, 1989). However, if the mare is a poor doer, or being ridden, her energy intake may need to be supplemented. This may be achieved in theory by an increase in hay intake. However, in late pregnancy, the increase in uterine size begins to limit the capacity of the digestive tract. Energy levels of $19.7 \text{ Mcal day}^{-1}$ ($2.40 \text{ Mcal kg}^{-1}$ (100% DM)) are required during this period. Good-quality feeds, low in bulk but high in nutrient value, are advised. In late pregnancy, the mare's roughage intake should therefore be reduced and partly replaced by increasing levels of energy-rich concentrates. Care should be taken to ensure that the required protein intake is still maintained (Tables 14.2–14.5; National Research Council, 1989; Pugh and Schumacher, 1990; Frape, 1998).

14.5.2.3. Vitamins and minerals

Vitamins and minerals are classified as micronutrients. The specific effects of deficiencies in many micronutrients on the pregnant mare and, indeed, on general equine welfare are as yet unknown. However, the importance of calcium (Ca), phosphorus (P)

and vitamin A is appreciated. The Ca:P ratio is of special importance and the involvement of both minerals in bone growth is well documented (Frape, 1998). In the pregnant mare, these micronutrients are important not only to the mare herself, but also to the fetus. Ca and P are normally stored within the bones, much of which are a temporary store and can be mobilized to satisfy demands elsewhere. If the pregnant mare's dietary intake of Ca or P is inadequate, especially during late pregnancy, the mare will mobilize her own stores from within her bones to satisfy the fetal demand. If the dietary deficiency is great, then her bones will suffer, become brittle and possibly be unable to take the strain of the increased weight in late pregnancy or the stresses of parturition. The foals of such mares can also suffer from deformities in tissue and bone growth and general ill thrift at birth.

The levels of Ca and P required in the first 8 months of pregnancy are of the order of 20 and 14 g day^{-1} (0.24 and 0.17% (100% DM)), respectively. During the last 90 days, the demand increases from 35 and 26 g day^{-1} (0.43 and 0.32% (100% DM)) in month 9 to 37 and 28 g day^{-1} (0.45 and 0.34% (100% DM)), respectively, at term. In the tenth month 25.3 mg Ca kg^{-1} of the mare's body weight are deposited in the fetus daily (Tables 14.3 and 14.4; National Research Council, 1989; Frape, 1998).

Not only are the absolute levels of these two minerals important but also their relative amounts. Excess P interferes with Ca absorption and leads to an effect similar to that of Ca deficiency. A ratio of Ca:P of between 1:1 and 6:1 is considered acceptable. Legume hay is a good source of Ca and P and supplementation of these minerals should not be required for mares fed legume hay *ad libitum* (National Research Council, 1989; Hintz, 1993a). It is important to know that straights in the form of grains tend to be relatively high in P and, therefore, in late pregnant mares, when increasing concentrate intake, Ca should be supplemented if grains are fed. Ca may be supplemented in the form of bone-meal (not so popular today), ground limestone flour or milk pellets, which are

more readily available in today's market. The high concentration of P in bran largely precludes its use in late pregnancy, except in small quantities as a laxative.

Other minerals should also be supplemented, though their exact function is unclear: for example, salt, potassium, magnesium, zinc, cobalt, iodine, manganese, copper, etc. (Ott and Asquith, 1986; Ott, 2001). These may be easily supplemented by one of the commercially available vitamin and mineral blocks, providing the mares with free access (Tables 14.3 and 14.4).

Vitamin A, as mentioned, is also important, especially as it is an essential component of epithelium. As such, it is important in reproductive function, cell regeneration and development. Adequate vitamin A levels are best ensured by feeding fresh green forage (Table 14.2); mares with no access to fresh pasture must, therefore, be supplemented (Frape, 1998). Excess supplementation as well as deficiency of minerals and vitamin A has been implicated in abnormalities associated with bone growth, such as developmental orthopaedic disease, angular limb deformities and epiphysitis (Beard and Knight, 1992).

14.5.2.4. Water

As with all equine rationing, water is an essential, but often forgotten, component. The pregnant mare requires large amounts of water, approximately 50 l day⁻¹, depending on the DM content of her ration (Huff *et al.*, 1985). This water must be clean, fresh and available at all times to allow consumption in small but frequent amounts.

14.5.3. Parasite control

Parasite control is of significant importance. A high parasite count is often the reason why some mares appear as poor doers, and parasites cause a large quantity of the food fed to be wasted. If a mare's internal worm burden is excessive, the damage caused may become permanent, condemning that mare to being a

bad doer for the rest of her life, or may even be a cause of death. Strongyles and ascarids are the main parasites associated with disease in mares and foals (Shideler, 1993d). Pinworms, tapeworms and bots may be present but rarely result in life-threatening conditions (Card, 2000). As with all equids, worming should take place regularly – at least every 12 weeks – during spring and summer. Less frequent worming is required in winter once the frosts appear; if temperatures do not fall below freezing, continuous 12-week worming throughout the year should be practised.

The development of resistance to wormers (anthelmintic treatments) can be avoided by rotating the wormer types used, i.e. those based on the thiabendazole group, followed by those based on the pyrantel embonate group. Care must be taken in worming pregnant mares, as not all wormers are suitable. Products based upon benzimidazoles, fenbendazole, pyrantel pamoate and ivermectin are considered safe (Varner, 1983; Card, 2000), but those based on cambendazole, for example, are not suitable for very early pregnant mares, within the first 12 weeks. Their use has been associated with a risk of abortion. At the other end of pregnancy, organophosphate wormers used to control bot flies are not recommended, as they may disrupt and trigger smooth-muscle contraction and may induce late abortions due to uterine contractions. In general, it is recommended that no wormers be used in the last month of pregnancy, due to the risk of inducing premature delivery (Varner, 1983). However, some advocate worming immediately prior to parturition to ensure that the foal is not exposed to a high parasite burden at birth (Shideler, 1993d; Card, 2000). The importance of clean grazing must not be overlooked and a combination of worming treatment, clean grazing and manure removal gives the best results (Herd, 1987a,b).

Delousing powder may be administered to pregnant mares, but its use should be avoided in late pregnancy. Ideally, pregnant mares should not be allowed to get into such a condition that such treatment is required.

14.5.4. Vaccinations

The vaccination programme required by a mare depends upon the endemic diseases prevalent and hence the country in which she lives. In the UK, vaccination against tetanus and influenza is automatic and is normally administered 4 weeks prior to parturition to allow the mare's titre of antibodies to be raised adequately to ensure transfer to the colostrum and hence to the foal immediately post-partum (Varner, 1983; Pugh and Schumacher, 1990; Robinson *et al.*, 1993).

Equine rhinopneumonitis (equine herpesvirus type 1 (EHV1)) is a problem in the USA and is becoming increasingly so in UK. This infection causes abortion, normally in the last trimester, in up to 70% of infected mares. If an outbreak is suspected or routine protection is required, vaccination can be administered in months 5, 7 and 9 of pregnancy (Witherspoon, 1984; Mumford *et al.*, 1996).

Equine viral arteritis (EVA) also causes abortion and a modified live vaccine is now available. Although EVA is a problem in parts of Europe and USA, it is not common at present in UK, but complacency cannot be allowed. Vaccination is available and may be given to pregnant mares but not in the last 2 months of pregnancy (Timoney and McCollum, 1997).

In other parts of the world, routine vaccination for eastern and western equine encephalomyelitis, equine pneumonitis (EHV4), rabies and Potomac horse fever may be considered. With the exception of rabies vaccination, all can be administered in the last 4 weeks of pregnancy to confer protection on the foal via colostrum. Vaccination for strangles, botulism, anthrax, *Salmonella*, Typhimurium and leptospirosis is possible but only advised in areas of

particular risk (Wilson, 1987; Sprouse *et al.*, 1989; Card, 2000).

14.5.5. Teeth care

The teeth of the pregnant mare should not be neglected. Regular teeth rasping ensures that all the plates are level and can efficiently grind food during mastication, enhancing digestion and maximizing the nutrient value of the food. This is of utmost importance when her system is under stress – for example, during late pregnancy.

14.5.6. Feet care

The majority of brood mares are unshod; even so, regular filing should occur. Poor feet cause pain, and this pain can be exacerbated with the increased weight burden of late pregnancy. Such mares will be reluctant to exercise themselves, resulting in problems, as previously discussed. Some mares are turned to stud with musculoskeletal problems and, as such, may need orthopaedic shoeing, especially in late pregnancy. Even so, shod mares must have shoes removed prior to parturition to prevent accidental damage to the foal.

14.6. Conclusion

Accurate pregnancy detection is a major key to early pregnant-mare management. Once pregnancy has been confirmed, mare management can be geared towards ensuring that pregnancy is stress-free, optimizing the chances of a healthy foal and a mare in the appropriate condition for lactation and subsequent re-covering.

15

Management of the Mare at Parturition

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15.1. Introduction

Gestation in the mare lasts on average 330–336 days, though considerable variation is evident (Bos and Van der May, 1980; Rossdale and Ricketts, 1980; Badi *et al.*,

1981). The physical process and endocrine control of parturition have already been detailed in Chapters 7 and 8. This chapter will consider solely the management of the mare at parturition, including its artificial induction and dystocia.

15.2. Pre-partum Management

Approximately 6 weeks before the mare's estimated date of delivery, she should be introduced to the foaling unit or the yard at which she is to foal. This begins the gradual familiarization of the mare to the surroundings in which she will foal and be kept immediately post-partum, so reducing the stress of any sudden changes. Such familiarization allows her to become accustomed to particular management practices, especially if she is to foal away from home. Changes in feed, exercise, housing and routine can be introduced in plenty of time to allow a regular management routine to be developed prior to foaling.

If the mare is to foal away from home, a period of 6 weeks is also required to allow her immune system to raise the necessary antibodies against any challenges present in her new foaling environment. This will not only provide protection for the mare herself, but will also allow the antibodies to pass into the colostrum and so provide the foal with immediate protection at birth. Bearing this in mind, it is advised that every mare should be vaccinated with either her annual booster or a new vaccination programme during this 6-week period. The vaccinations used depend upon the country of residence, prevalent diseases and types and ages of mares, as discussed in Chapter 14 (Golnik, 1992; Card, 2000).

Exercise is very important. Regular free exercise in a paddock or field will be adequate for most mares and will help maintain their fitness for foaling and reduce the chances of oedema (fluid retention) in the legs. A slightly laxative diet may be advised, as many mares suffer from constipation in late pregnancy, especially if exercise is limited. To this end, components such as bran may be added in small quantities, but care must be taken not to upset the overall nutritional balance of the diet, especially the calcium:phosphorus (Ca:P) ratio, which is very important at this stage. Lastly, but by no means least, clean and fresh water must be available at all times. Mares need large quantities of water in late pregnancy (Frape, 1998).

Throughout the preparation of the mare for foaling, she should be observed for the characteristic signs of imminent parturition: increase in mammary-gland size; the secretion of milk; and general relaxation of the abdominal, pelvic and perineal area (see Chapter 7).

By this time, except in the case of an emergency, it will have been decided whether the mare is to give birth naturally or if birth is to be induced. If birth is to occur naturally, then, as soon as any signs of imminent parturition are noticed, the mare must be put into her foaling box, if she is to foal inside, or into a small quiet paddock, if she is to foal out. She should then be monitored closely.

If she is to foal inside, the box provided must be at least 5 m × 5 m, with good ventilation, but draught-free. Traditionally, the floor covering would have been a deep bed of straw, which provides a soft, warm, dust-free surface on to which the foal can be born (Fig. 15.1.). More recently, rubber matting has become increasingly popular (Fig. 15.2). Such matting is expensive but provides a clean, insulated and dust-free floor that can be easily washed and disinfected.

The foaling box should be free of any protrusions, which might cause damage to the mare or foal. Ideally, it should have rounded corners to reduce the risk of the mare getting cast. Hay nets should not be used, as the foal can get itself caught up in anything left dangling. Hay should be fed off the ground. The use of high hay racks avoids the wastage of feeding off the floor but does run the risk of the mare and foal getting seeds in their eyes and ears. In an ideal purpose-built unit, each box should have two doors, one to the outside for horse access and another facing into a central sitting area for human access and viewing (Fig. 15.3). Closed-circuit television is also a good method of viewing mares with minimal disturbance. The provision of radiant-heat lamps in each box is an advantage for weak foals.

Once the mare has settled into her foaling quarters, it is a case of careful watching and patient waiting. Careful observation can minimize the time from the first signs of trouble to action and can therefore be crucial in saving lives.



Fig. 15.1. The traditional floor covering for the foaling box was straw, which provides a good, deep, soft bed for the foal to be born on to.



Fig. 15.2. Rubber matting as a flooring for foaling boxes is becoming increasingly popular. It is dust-free and warm and can easily be hosed down and disinfected.

15.3. Induction of Parturition

The mare shows a much wider variation in gestation length than other farm animals in which induction of parturition is practised. As discussed in Chapter 8, the natural length of gestation is affected by several factors. When considering artificially inducing parturition, it is essential that the date is as close as possible to the estimated delivery date. Premature induction will result in foals with all the classical symptoms of prematurity,

including breathing difficulties, being late to stand and a delay in the normal post-partum adaptation mechanisms. Such foals may, if they survive, suffer long-term ill effects and there will be a dramatic increase in labour and veterinary expense. Induction of parturition in horses should therefore be carried out with great care and its value as a routine technique is questionable.

Induction may be carried out as an aid to management, to ensure that all facilities and staff are available and ready when required.



Fig. 15.3. An ideal arrangement of the foaling unit allows the foaling boxes to be viewed from a central sitting area (National Stud, Newmarket).

It is particularly useful if limited experienced labour is available and only a few mares are involved. Alternatively, induction may be used as a matter of urgency in the case of prolonged gestation, preparturient colic, pelvic injuries, ventral rupture, previous premature placental separation, pending rupture of the prepubic tendon or painful skeletal or arthritic conditions, which can get unbearable in late pregnancy. Finally, it may be used as a research tool.

15.3.1. Fetal maturity

As indicated, the timing of induction in relation to the expected natural delivery date and fetal maturity is crucial with regard to fetal/foal survival. In horses, parturition is related to fetal development, especially maturity of the adrenal cortex and its ability to secrete corticosteroids. In the 5 days preceding natural parturition, fetal cortisol levels are seen to increase significantly, and they are required for the final maturation of major organ systems (Rossdale *et al.*, 1997). In order for the fetus to survive parturition, it must have been exposed to these elevated cortisol levels; hence induction of parturition prior to this would not yield a viable foal (Chavatte *et al.*, 1997b). As gestation lengths are so variable in mares, it is not possible to use these

as more than a rough guide to determine the expected parturition date; hence fetal maturity needs to be determined by an alternative means. The normal signs of parturition (see Chapter 7) can again give an indication but are not accurate enough to use to time artificial induction (Le Blanc, 1997). However, changes in secretions of the mammary gland are a good indicator. Ca concentrations ≥ 4.0 mg ml⁻¹ are reported to indicate a mature fetus, while concentrations ≤ 1.2 mg ml⁻¹ indicate an immature fetus (Leadon *et al.*, 1984; Ousey *et al.*, 1984; Ley *et al.*, 1993). An inverse relationship between sodium (Na) and potassium (K) is also evident. As parturition approaches Na concentration declines and K concentration increases. When Na < K, then the induction of parturition is reported to be possible. Based upon this, Ousey *et al.*, (1984) devised a scoring system to determine when induction of parturition is safe (Table 15.1).

Haematological assessment of the fetus has also been suggested as an accurate means of determining the safety of induction (Jeffcote *et al.*, 1982; Rossdale *et al.*, 1984), as well as rectal palpation, vaginal examination, amniocentesis, fetal electrocardiography, fetal eye diameter and aortic diameter (determined by ultrasonography), but, when compared with the scoring system, they all have limitations (Reef *et al.*, 1995; Le Blanc, 1997).

Table 15.1. Scoring system used to determine when the induction of parturition is safe (from Ousey *et al.*, 1984). A total score of ≥ 35 suggests that safe induction of parturition is possible.

Colostrum concentrations			Points for each electrolyte
Calcium (mg dl ⁻¹)	Sodium (mEq l ⁻¹)	Potassium (mEq l ⁻¹)	
≥ 40	≤ 30	≥ 35	15
≥ 28	≤ 50	≥ 30	10
≥ 20	≤ 80	≥ 20	5

15.3.2. Methods of induction

Once fetal maturity has been determined, there are several methods that may be employed to induce parturition (Hillman and Lesser, 1980); these will be discussed in turn.

15.3.2.1. Corticosteroids

In ruminants, the most successful means of inducing parturition is the use of corticosteroids (Adams and Wagner, 1970). Foaling can be similarly induced in large mares using dexamethasone, a corticosteroid administered daily for 4 days from day 321 of gestation. Parturition is reported to occur within 1 week and to result in live and healthy foals (Alm *et al.*, 1974, 1975). However, other workers, using pony mares, report less success, indicating stillbirths and placental retention (Rossdale and Jeffcote, 1975; Van Niekerk and Morgenthall, 1976; First and Alm, 1977). This apparent lack of success in mares when compared with ruminants is a likely indication of the differences in endocrine control of parturition (see Chapter 8).

15.3.2.2. Progesterone

Progesterone administration over a period of 4 days in late pregnancy and its subsequent withdrawal will induce parturition over approximately 1 week (Alm *et al.*, 1975). Such use of progesterone does not result in parturition in ruminants.

Both these methods have the disadvantage of being relatively inaccurate in the timing of

the reaction to their administration, which can be over a period of 1 week, and the rate of stillbirths resulting. They are, therefore, not used commercially. A more accurate and more immediate induction agent is required.

15.3.2.3. Prostaglandins

The use of prostaglandins gives a more immediate result than that of progesterone and corticosteroids. Fluprostenol, 250–1000 μ g administered intramuscularly, is sufficient to cause parturition within 2 h in a mare at full term. The mare will show the initial signs of parturition (stage 1) within 30 min. The foal is usually born within 2 h and the afterbirth appears 2 h later. Complications, such as insufficient cervical dilatation and hence rupture and decrease in foal viability, have been reported (Rossdale *et al.*, 1979a; Ousey *et al.*, 1984; Ley *et al.*, 1989). In the natural course of events, the rise in prostaglandin coincides with the delivery of the foal. Fluprostenol presumably imitates this. Its use too early, therefore, has little effect, as elevated levels at this time are out of synchrony with other endocrine changes (Alm *et al.*, 1975; Van Niekerk and Morgenthall, 1976; Cooper, 1979).

15.3.2.4. Oxytocin

A further agent – the one most commonly used today to induce parturition – is oxytocin. In the natural course of events, oxytocin concentrations rise markedly during the second stage of labour, causing the rapid uterine myometrial contractions associated with birth. Initial research work used high

levels of oxytocin (60–120 iu), but these were associated with cervical rupture and reduced foal viability, a likely consequence of the sudden onset of myometrial contractions, bypassing the natural more gradual build-up of events. More recently, low levels of oxytocin, < 20 iu, have been used, administered over time (3 h) and this results in parturition within 1 h (Pashan and Allen, 1979b; Pashan, 1982). The exact amount of oxytocin required depends on the endocrine balance within the mare at administration (Pashan, 1980). It is known that, in many mammals, there is a synergistic relationship between oxytocin and prostaglandins and that such an association does occur in mares. Oxytocin administration near term, therefore, results in an immediate release of prostaglandins, equivalent to the natural release. Oestrogen can be used in combination with oxytocin as an induction regime and is reported to aid cervical dilatation and hence ease the process of parturition (Hillman and Ganjam, 1979).

It cannot be emphasized enough that, if induction of parturition is being considered, accurate records of the mare's date of service and expected delivery date are very important, along with close observation for signs of parturition and a means of determining fetal maturity. Inappropriate use of induction agents has disastrous consequences. Most of the danger is to the fetus rather than the dam, though the risk to both increases with the asynchrony between induction and the natural course of events. Induction of parturition in mares remains a risky business and must be used bearing this in mind.

15.4. Management of the Mare at Parturition

Whether the mare delivers naturally or is to be artificially induced, her management should be very similar. The main difference is that the time of delivery with induction will be known and therefore preparation can be better timed and organized. A mare may foal in a specifically built foaling unit or outside. Whichever system is chosen, and there

are advantages in both, the principles of the stages of labour and their management will be the same. Foaling mares outside is increasingly popular, as the risk of disease is lower and the system is much nearer the natural situation. Foaling outside is normally restricted to pony- or cob-type hardy mares or multiparous mares foaling later on in the season. Early foalers, maiden or difficult mares and those of great value are normally foaled inside, allowing closer observation.

15.4.1. Foaling kit

In readiness for foaling the following equipment should be organized:

- Veterinary telephone number – ideally, the vet should also be warned beforehand
- Mare halter and lead rope
- Towels
- Bucket
- Soap or antibacterial wash
- Cotton wool
- Access to warm water
- Obstetric lubricant
- Obstetric ropes
- Sharp knife or scalpel
- Radiant-heat lamp
- Antiseptic spray/navel dressing – e.g. 0.5% chlorhexidine
- Feeding bottles for milk/colostrum
- Gastrointestinal feeding tube
- Access to colostrum, e.g. frozen colostrum

15.4.2. First stage of labour

First-stage labour requires no special measures except close observation for the start of the second stage and for the identification of any potential problems. Once first-stage labour has been diagnosed, the mare's tail should be banded up and the perineal area thoroughly washed. Some mares may lose milk prior to or during the first stage of delivery (see Fig. 7.2). This milk is valuable colostrum, of which there is a finite amount. If a mare shows considerable milk loss prior to parturition, she should be milked lightly and the colostrum collected into a clean sterile container. As soon

as the foal is born, this can be bottle-fed to the foal, or tubed, if necessary, to ensure that valuable antibodies are received.

During the first stages of labour, the mare will seem restless and may repeatedly get up and lie down. She may well sweat profusely and appear uneasy, glancing at her flanks and grimacing; she may also dig up her bedding.

This continual moving around is thought to help to position the foal within the birth canal. The mare may well show signs of discomfort, followed by quiet. Thoroughbreds are notorious for this type of behaviour (Jeffcote and Rosedale, 1979). Whatever her behaviour, her discomfort will increase with the frequency of contractions, culminating in the breaking of the waters (release of allantoic fluid) at the cervical star. Excessively prolonged first-stage labour may be a sign of problems, especially if the mare seems to be very distressed. It is very difficult to state how long first-stage labour should last and at what stage you should call for assistance, as some mares will naturally show several false starts in the days preceding birth. However, as a general rule, the assistance of a veterinary surgeon should be sought if the mare seems to be in prolonged discomfort, showing considerable agitation and profuse sweating, and before she is in any danger of becoming exhausted.

15.4.3. Second stage of labour

The management of the second stage of labour is more important and is marked by the breakage of the chorioallantois at the cervical star and the resultant release of allantoic fluid. In 90% of cases, the mare will now take up a recumbent position, the most efficient for straining (Fig. 15.4).

At this stage an episiotomy (cutting of the vulva and perineal area) should be performed, if required. Unfortunately, today many mares routinely undergo Caslick's operations (see Chapter 1), due to inadequate perineal conformation. Such mares, along with those that are naturally small, will need an episiotomy to allow passage of the foal (Fig. 15.5).

As soon as this has been done and within 5 min of the start of the second stage of labour in mares not requiring an episiotomy, a brief internal examination may be made to ensure that the foal is presented correctly (Fig. 7.8). At this stage, the amniotic sac should be evident as a white membrane bulging through the mare's vulva (Fig. 7.7).

If the foal's forelegs, one leg slightly in advance of the other, and muzzle can be felt within the vagina, the mare should be left alone to deliver naturally (Figs 7.9 and 7.13). If there are problems, assistance should now



Fig. 15.4. During second-stage labour the mare invariably takes up a recumbent position, the most effective for straining (photograph Steve Rufus).



Fig. 15.5. An episiotomy should be performed by the beginning of stage 2 labour. This is routinely required in the case of mares having undergone a Caslick's operation.

be called. Care should be taken not to rupture the amnion during this process. Ideally it should be left to break naturally. When it does the colour of the amniotic fluid should be noted for evidence of meconium staining – dark brown/green coloration – which is indicative of fetal stress. If this is the case, then delivery of the foal should be speeded up by traction as soon as the foal's head appears (Figs 7.11 and 15.6).

If all is well, all attendants should now leave the box and allow the mare to foal unaided but observed from a discrete distance. Most mares lie down during second-stage labour, as this is the most efficient position for voluntary straining. Plenty of room is required to allow the mare to stretch out fully during straining.

The vast majority of foalings, up to 90%, require no outside interference from humans (Vandeplassche, 1993). Occasionally, how-

ever, things do go wrong and it is as well to be prepared for, and have an understanding of, such eventualities. In such cases, prompt action can often save the life of both foal and mare. Foaling abnormalities are considered in detail at the end of this chapter.

Second-stage labour should last on average 15 min (range 5–30 min). Mares foaling for the first time tend to have a longer second stage-labour and so do mares that have had a hard first-stage labour.

15.4.4. Immediately after delivery

After delivery, the foal should be left with its hind legs still within its mother and the umbilical cord intact (Fig. 7.12). The umbilical cord must be allowed to break naturally to minimize blood loss (Rossdale, 1967; Fig. 15.7). The foal lying with its legs within the vulva of the dam appears to have a tranquillizing effect on the mare. As a result, most mares are reluctant to get up immediately, though they may turn to lick the foal. A mare may remain recumbent for 30 min post-partum. This should be encouraged, as it allows initial recovery of the tract and reduces the inspiration of air and therefore bacteria through the still relaxed vulva (Fig. 15.8). Such contamination of the vagina increases the chance of post-partum endometritis or acute metritis and delays uterine involution and any return to oestrus. Temporary Michel clips may be used after stage 3 to hold the dorsal (upper) vulval lips together and reduce air contamination. These clips can easily be removed at the normal day 2–3 post-partum veterinary examination. They can then be replaced with a Caslick operation, if required. This period marks the beginning of maternal foal bonding and recognition. Some mares, especially if stressed or a maiden, may get up and paw the ground at some danger to the foal.

If the foal has shown signs of distress and is limp or weak at delivery, the amnion should be broken immediately and the foal's head lifted to aid breathing (Fig. 15.9). Occasionally, the umbilical cord does not break after birth, despite drying up and constriction at the foal's abdomen. In such cases,



Fig. 15.6. Gentle traction may be used to aid in the final stages of delivery, especially if the mare is showing signs of exhaustion (photograph Steve Rufus).



Fig. 15.7. Very soon after parturition the healthy foal should be alert and sternally recumbent. The umbilical cord should be left to break naturally. This normally occurs as the foal makes its first attempts to rise to its feet (photograph Steve Rufus).

it may be broken by a sharp pull while placing the other hand on the foal's abdomen.

Immediately after delivery, the foal can be dried off (Fig. 15.8) and the severed umbilical cord must be dressed with an antiseptic agent such as 0.5% chlorhexidine to prevent infection. Iodine was traditionally used as a navel dressing. This is effective in preventing infection, but may cause sloughing off of skin cells, causing sores and the possible reopen-



Fig. 15.8. The foal may be dried off immediately after delivery. At the same time the heart rate of the foal can be checked by placing a hand on the side of the foal's thorax. The mare should be encouraged to remain recumbent for a while after birth in order to reduce the inspiration of air, and, therefore, bacteria in through the relaxed vulva (photograph Steve Rufus).

ing of the site. The milder, but just as effective, chlorhexidine is now advised. At this stage, the foal's heart rate may be checked by placing a hand on the thorax (Fig. 15.8). It



Fig. 15.9. The amnion may be removed from the foal's head and the nasal passages cleared to aid initial breathing (photograph Steve Rufus).

may also be weighed, the birth weight of most foals is 10% of their expected mature weight. Details of the parameters required to be met by the foal at set stages in the first few hours of life are given in Chapter 16.

15.4.5. Third stage

During third-stage labour, the mare will appear restless again, similar to that during the first stage. The average duration of this third stage is 60 min but there is wide variation. Occasionally, the placenta may be expelled immediately after, or with, the foal and still attached to the umbilical cord. At the other extreme, it may take several hours. If the placenta is not expelled before the mare stands, it may be tied up to prevent the mare standing on it and ripping it out prematurely. The extra weight provided by tying up also encourages its expulsion (Figs 15.10 and 15.11). Third-stage labour in excess of 10 h is indicative of problems and will be discussed in the following section.

As soon as this third stage has been completed and the placenta expelled, it should be removed from the box and examined for completeness (Fig. 7.14). An effective method of detecting holes and therefore any missing fragments is to tie off both uterine horn ends of the placenta and fill the placenta with water through the cervical star. Leakage of water indicates a break, which

should be examined to ensure that no membranes have been retained. If this is suspected, then a veterinary surgeon should be called to ensure their complete removal. Placental retention, even of just a fragment, can lead to acute metritis, septicaemia and eventual death if not treated as a matter of urgency (Chapter 19). The temptation to pull the placenta to try and release it must be resisted, as this will invariably lead to



Fig. 15.10. The allanto-chorion (placenta) can be tied up to prevent the mare standing on it and ripping it out prematurely or partial placental retention (photograph Steve Rufus).



Fig. 15.11. Uterine contractions of the third stage of labour plus vasoconstriction of the placental blood vessels should result in expulsion of the placenta. Accidental tearing of the membranes can lead to partial placental retention, which must be treated immediately (photograph Steve Rufus).

rupturing of the placental membranes and the danger of fragments being retained. Only a very small fraction is required to set up a septicaemia infection.

During this period, the bond between mare and foal starts to develop and can be irretrievably damaged by human interference, however well intentioned (Fig. 15.12). Damage to newborn foals by their mothers is very rare, and if it does occur, it is usually a result of her being disturbed or flustered by unwanted human interference. Occasionally, mares will nibble or gently bite their foals to encourage them to move. Within reason, this may be allowed, but, if the dam is too aggressive, a muzzle may be used. Very occasionally, usually in maiden mares, real aggression towards the foal may be evident, involving kicking and vicious attack. Such mares may be tranquillized for a short period of time.



Fig. 15.12. During the third stage of labour the mare and foal should be left undisturbed to start the process of bonding. There is no need to drag the foal around to the head of the mare as she will invariably reach around herself and move towards the foal without assistance (photograph Steve Rufus).

Tranquillizers may also be used on mares that will not stand to allow the foal to suckle. After a while, most mares will get used to the foal and interference will not be required. Use of the twitch should be avoided for the first 24 h post-partum, as this is thought to increase the risk of internal haemorrhage.

Foals that appear to be weak and unable to get adequate colostrum from the mare may be bottle-fed or tubed. This ensures that they receive sufficient of the essential antibodies as soon as possible.

Enemas used to be routinely administered to foals, using either medical paraffin or warm soapy water. This treatment is no longer deemed necessary unless the foal shows signs of meconium retention after 24 h. Foals should be given the opportunity to suckle and pass meconium naturally before the decision is made to use an enema. The enema technique causes the foal unnecessary stress and rough handling at a very early age when delicate adaptation to the extrauterine environment is still occurring.

The mare should be given a feed about an hour after the birth. A light, easily digested and slightly laxative feed is best, plus fresh hay and water. Water should be given only under supervision initially, unless automatic water feeders are installed, to minimize the risk of the foal drowning.

15.5. Foaling Abnormalities

Foaling abnormalities or dystocia can be classified as a stress or complication associated with parturition that prevents a natural birth. It can be divided into fetal dystocia, resulting from fetal complications, or maternal dystocia, resulting from maternal problems. Dystocia often causes a delay in parturition, which may result in a reduction in the oxygen intake by the foal, due to partial placental breakdown or constriction of the umbilical blood supply. This may result in the birth of a weak foal, requiring careful intensive postnatal care and/or more permanent problems, including brain damage and death in the more extreme cases. Delay in parturition due to dystocia may result in other complications, further hampering progress. The uterus may close around the fetus as it dries out and the lubricating effect of the allantoic fluid diminishes. As a result, manipulation of the foal becomes increasingly difficult. Further drying out of the tract due to delay hinders passage through the birth canal and thus makes any successful manipulation even harder.

If dystocia does occur, there are three main actions that can be taken: manipulation and traction; caesarean; and fetotomy. Initially, the least drastic option of manipulation, or mutation, of the foal (manual correction of the fetal position), followed by traction, or pulling, of the foal should be considered. However, more drastic action may be required, such as a Caesarean, especially in cases where extensive manipulation would be needed. A Caesarean is a higher risk to both mare and foal and must be conducted under some form of anaesthetic. Mortality rates for foals born by Caesarean are approximately 10%. The mortality rates

for mares delivering by Caesarean are lower than this, especially if she is operated upon before labour has progressed too far (Vandeplassche *et al.*, 1979a). A Caesarean involves either a ventral midline incision, with the mare in a recumbent position, or a flank incision, if she is standing. Apart from the mortality risk, mares that have undergone Caesareans have the added risk of developing adhesions. Adhesions form between internal structures that have become damaged, often by exposure to air. If the adhered structures include parts of the reproductive tract, there is a risk of interference with their function. Infection is also another potential problem, though with the advances in modern medicine this is nowhere near the risk that it used to be (Vandeplassche *et al.*, 1979a). A final alternative, in response to severe cases of dystocia, is fetotomy. This is performed on foals that have died *in utero* and involves the dividing up of the foal's body within the mare. A Caesarean is a possibility in such cases but involves a higher mortality risk to the mare. Therefore, if the foal is already known to be dead, fetotomy is often the preferred option.

15.5.1. Fetal dystocia

Fetal dystocia is caused almost exclusively by malpresentations *in utero*, making normal unaided birth impossible. The foal *in utero* may be found in one of many positions, and the ease of correction and likely outcome depend on its position and the skill of the person manipulating it (Vandeplassche, 1993; Blanchard, 1995). Some of the more common conditions and their treatment are given below.

15.5.1.1. Forward positions

One or more forelegs may be flexed or folded under the foal whose head is in the normal position within the birth canal. As discussed previously (section 7.3.2), the widest cross-section presented is the thorax area; therefore, in this position, it includes the flexed forelegs, making passage impossible (Fig. 15.13).

Fig. 15.13. Flexion of one or both forelegs at delivery significantly increases the cross-section across the foal's thorax and therefore makes the passage of the foal very difficult.

The head of the foal may be flexed back. In this position, the nose of the foal will not be felt in the birth canal. This again presents a much wider maximum cross-section than the mare can deliver naturally (Fig. 15.14).

The foal's legs may be more misaligned than the normal hoof-to-fetlock alignment. One leg may become caught on the pelvic brim at the point of the elbow, preventing the foal's delivery (Fig. 15.15).

The foal's forelegs may lie over its head and be lodged behind its ears. This presents a cross-section too large for natural passage and runs the risk of rectal vaginal fissure if the hoof penetrates the roof of the vagina and into the rectum (Fig. 15.16).

All four of these positions can, in theory, be corrected reasonably easily by pushing the foal back into the uterus and manipulating it so that it is presented in a normal position, followed by traction to aid the mare in its delivery. In practice, the strength of the uterine contractions can make this quite difficult. It must also be remembered that traction has to be applied in a curved manner to ease the foal along the birth canal, as dictated by the pelvic anatomy.

Other more complicated positions are seen, which require veterinary assistance, and, in the worst scenario, a Caesarean may be required, especially if straining by the mare has caused damage to the cervix, vagina or uterus. The foal may have both its head and neck turned back, presenting just two forelegs (a more extreme version of that shown in Fig. 15.13), or the presentation of the head and forelegs is correct but the hind legs are also being presented at the same time (Fig. 15.17), putting four legs in the birth canal. Four legs are also presented first in the crosswise position (Fig. 15.18).

Finally the foal may be presented in a ventral position, in which case the arch of the foal's spine does not allow expulsion in the required curved manner. This position must be corrected by rotation of the foal before delivery is possible (Fig. 15.19.)

15.5.1.2. Backward presentation

Backward positions may be evident, with the back legs presented first, a position that need not be manipulated *in utero* but must be carefully watched during labour. In such cases, there is a danger that the umbilical cord may

Fig. 15.14. If only the forefeet are presented in the birth canal the head may be flexed back, again presenting such a wide cross-section of foal that it is very difficult for the mare to foal naturally.

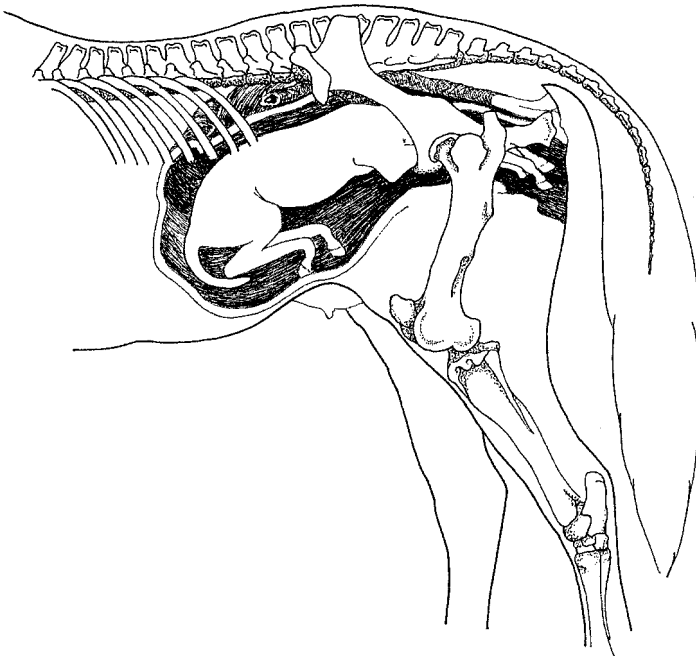


Fig. 15.15. A significant misalignment of the hoof of one leg and the fetlock of the other may be due to one elbow being flexed and becoming lodged at the pelvic brim.

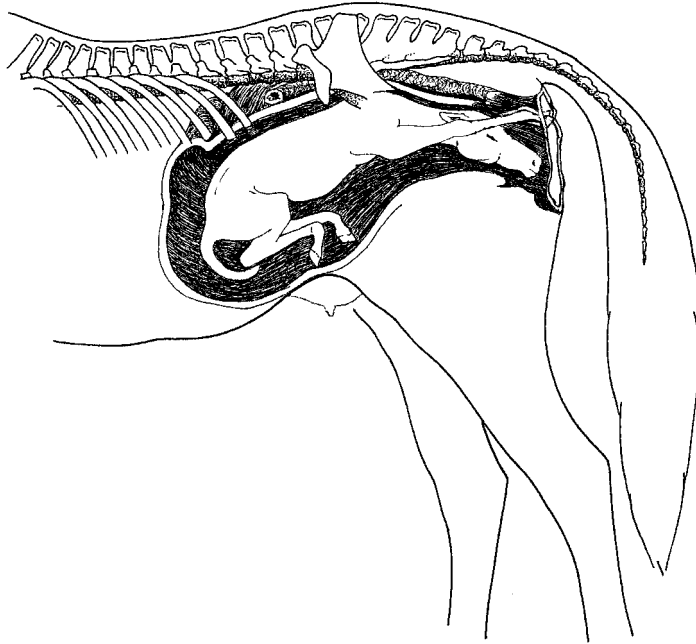


Fig. 15.16. The forelegs may be positioned over the foal's head. Not only does this increase the cross-sectional diameter of the foal but it also creates the risk of a rectal vaginal fissure.

Fig. 15.17. If both forefeet and hind feet are felt within the birth canal, veterinary assistance should be called immediately, as this position can prove very difficult to correct, especially if the mare has progressed far into labour.

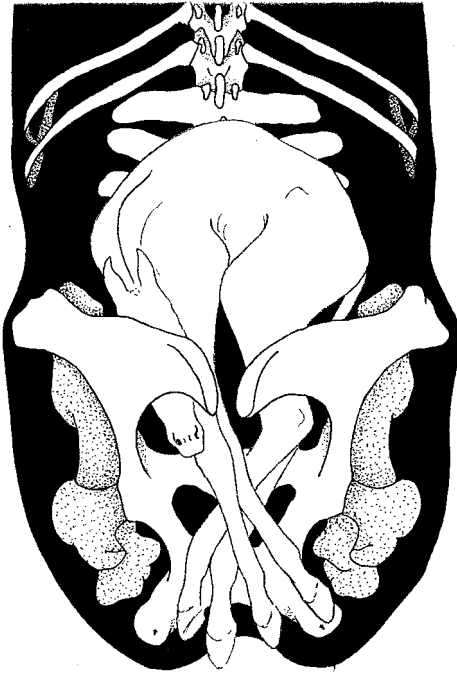


Fig. 15.18. A crosswise presentation also presents four feet first. This can, in theory, be corrected by pushing the hind legs back into the uterus.

become trapped between the abdomen of the foal and the pelvic brim, starving the foal of oxygen. There is also the danger that the foal may drown by inspiring allantoic fluid *in utero*. Foals in this position must, therefore, be pulled quickly and the amniotic sac removed from the muzzle to allow extrauterine breathing as soon as possible.

A more complicated backward position is indicated when no back legs are presented and only the tail can be felt. This position is known as a breech and veterinary assistance will be required to deliver such a foal (Fig. 15.20).

Other positions, variations of the above, may be found; those discussed are those most commonly observed.

15.5.2. Maternal dystocia

Maternal dystocia is the failure of natural delivery as a result of a maternal complication. These complications may be found in isolation or in combination (Blanchard, 1995). Some of the more common ones will be discussed.

Fig. 15.19. Ventral position of the foal. The foal must be rotated into a normal position before delivery is possible.

Fig. 15.20. If no legs can be felt within the birth canal and only the tail and rump is presented, this is a true breech position and will require veterinary assistance.

Placenta praevia is evident as the protrusion of the intact red allanto-chorionic membrane through the mare's vulva, due to its failure to rupture at the cervical star. This may be due to extra thick placental membranes, in which case manual breakage of the protruding allanto-chorion will allow parturition to progress normally. However, failure of the allanto-chorion to rupture at the cervical star may be due to rupture elsewhere, indicating premature separation of the placenta from the maternal epithelium. As this separation is one of the triggers for the foal's first breath, there is a danger of suffocation. The condition may also be indicative of placentitis, which, if long-standing, is likely to have led to fetal brain damage (Vandeplasse, 1980, 1993).

Small pelvic openings may also cause problems. Restriction in this area may be the result of an accident or fracture or caused by malnutrition during the mare's early developmental stages of life. In such cases, a Caesarean is often the only course of action that can be taken. It is questionable whether such mares should be used for breeding.

Uterine problems may also cause dystocia. One of the more rare conditions is uterine torsion, where the uterus has become twisted, often as a result of a fall or weakened broad ligaments. The twist, which is often towards the cervix end of the uterus, prevents any delivery through the birth canal. The prognosis for such cases is normally poor, but some success has been reported by enlisting the help of gravity and rolling the mare over rapidly or by using manual correction via a flank incision or via the rectum. Often a Caesarean is required (Vandeplasse, 1980; Pascoe *et al.*, 1981). Uterine inertia is a more common condition and may often accompany fetal dystocia. In such cases, the uterine myometrium becomes exhausted due to excessive straining, especially during the second stage of labour. The condition can be accentuated by age or previous uterine infections and may result in uterine rupture or placental retention. Uterine inertia may not occur to the same extent across all the myometrium. In such cases, tight rings of muscle contraction may occur. As a result, there is a danger that

the foal may be crushed or strangled. In most cases of uterine inertia, traction to aid the mare, along with an oxytocin injection to encourage myometrial contraction, is all that is required. Occasionally a Caesarean may be advised (Vandeplasseche, 1980, 1993).

15.5.3. Abnormal conditions

Abnormal conditions associated with parturition do not normally directly prevent normal delivery but may indirectly cause concern (Lofstedt, 1993; Blanchard, 1995). These conditions include problems that become evident prior to labour, such as ruptured prepubic tendon, uterine prolapse and uterine torsion. The prepubic tendon is the tendon sheet attached to the abdominal muscles and so supports the entire abdominal contents, including the uterus. Its rupture results in loss of support for the abdominal organs and death is the normal outcome (Fig. 15.21; Jackson, 1982).

Uterine prolapse results in the inversion of the vagina, and often part of the uterus through the vulva, due to incompetent uterine support, possibly associated with old age. In such cases, the uterus can be replaced manually after the administration of an anaesthetic or relaxant. This reduces the

mare's straining and allows the uterus to be replaced and the vulva to be sutured to prevent a recurrence (Brewer and Kleist, 1963; Vandeplasseche, 1975; Breen Vann and Bowman, 1994).

Uterine torsion, as previously discussed, is the term given to a twisted uterus, often due to stretched broad ligaments. The twist prevents the natural passage of the foal at parturition (Bowen *et al.*, 1976).

Some conditions do occur during parturition itself – for example, uterine or intestinal rupture. Uterine rupture most commonly occurs at parturition due to: mutation or traction; excessive straining, especially during second-stage labour; or significant intrauterine movement (Wheat and Meager, 1972; Patel and Lofstedt, 1986). Intestinal rupture is associated with the feeding of large, infrequent meals during late pregnancy (Littlejohn and Ritchie, 1975; Fisher and Phillips, 1986). Both uterine and intestinal rupture may be the result of a weakness arising from previous damage to these organs.

Finally, some conditions may not become evident until after parturition but are a result of the forces of delivery. For example, haemorrhage, both internal, often the cause of sudden death post-partum, and the less drastic external, may be a result of labour but not detected until after delivery (Rooney,



Fig. 15.21. Prepubic tendon rupture may occur in late pregnancy, resulting in the loss of all abdominal support. The prognosis is not good; death normally results (photograph Julie Baumber).

1964; Pascoe, 1979b). Rectal vaginal fissure or perineal lacerations may be evident, resulting from the foal's hoof puncturing the roof of the vagina and passing through the floor of the rectum, opening up a cloaca as the foal is delivered (Fig. 15.22). Short fissures may be sutured, but longer ones cause significant problems, largely due to inaccessibility.

Retention of the placental membranes may cause serious complications. Normally, the placenta reduces in size gradually, as its blood flow reduces; it therefore shrinks and separates away from the endometrium and is delivered by the final contractions of the uterus in third-stage labour. Retention of the placental membranes will result in infection, which, if left untreated, will prove fatal. Retention of the afterbirth for more than 10 h indicates problems. Retention is normally in the previously non-gravid uterine horn, especially after dystocia or Caesarean section. It may be due to a hormonal imbalance, resulting in inadequate oxytocin release and therefore reduced muscle activity. Treatment can be in one of several ways, including manual removal

and/or oxytocin treatment. Manual removal must be carried out with great care and, if attempted, it must be certain that the entire placenta is removed. After initial attempts, an antibiotic pessary can be inserted and the rest of the membranes removed a few days later. It is imperative that the entire placenta is removed as soon as possible. If the whole of the placenta has been retained, then iodine solution can be pumped in through the cervical star to fill up the allantoic sac. It may take 9–11 l for an effect to be obtained. After about 5 min, the mare will be seen to strain against the filled placenta and so aid expulsion. This method can be used in conjunction with oxytocin treatment, though the pressure on the uterus itself induces an elevation in endogenous oxytocin (Threlfall, 1993; Haffner *et al.*, 1998).

Delayed involution may be apparent after parturition. Under normal conditions, within a few hours the uterus should have shrunk to one-quarter of its fully expanded state. By day 7 post-partum, it should be only two to three times the size evident in a barren mare,

Fig. 15.22. During foaling the foal's feet may pass through the vagina wall and up into the rectum, forming a rectal–vaginal fissure.

returning fully to its normal size by day 30 post-partum. Evidence of a dilated cervix or enlarged uterus at day 30 indicates problems, possibly associated with retention of placental membranes or dystocia. Uterine involution can be encouraged in such mares by daily infusion of oxytocin and antibiotics, plus gentle exercise (Blanchard and Varner, 1993a). Exogenous oxytocin may be used to provide the extra impetus for placental expulsion followed by uterine involution (Threlfall, 1993; Haffner *et al.*, 1998).

Finally, hypocalcaemia, though not common in mares, if evident can be quite successfully treated by administration of calcium borogluconate post-partum. The

incidence of hypocalcaemia is higher in mares suffering from dystocia and after Caesarean section.

15.6. Conclusion

Management of the foaling mare is of utmost importance in ensuring the birth of a healthy foal. Inappropriate management or the failure to call in professional help when required can have disastrous consequences. Induction may be used as an aid to the management of the foaling mare, but must be used with great caution, as inappropriate timing can have fatal consequences.

16

Management of the Lactating Mare and Young Foal

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16.1. Introduction

Correct management of the lactating mare and young foal is crucial for long-term foal survival and the rapid return of the mare to oestrus with successful re-covering. As far as the foal is concerned, the adaptation from intra- to extra-uterine environment is the most crucial stage. The subject of neonatal complications and disease is vast and beyond the scope of this book. This chapter will therefore concentrate on the normal foal in order that abnormal foals can be identified. More specific detailed texts should be consulted on the problems that may be encountered in the neonate (Koterba, 1990; Madigan, 1990; Adams, 1993a,b,c; McClure, 1993; Reef, 1993; Roberts, 1993; Seltzer *et al.*, 1993; Traub-Dargatz, 1993a,b; Vaala, 1993; Welsch, 1993).

16.2. Foal Adaptive Period

Immediately post-partum, the foal has to undergo substantial anatomical, functional and biochemical adaptive changes in order to survive in the extra-uterine environment. In the normal foal, adaptive changes can be identified until puberty and even up until the achievement of mature size. However, in considering the true adaptive period for survival outside the uterus, the first 4 days of life are the most crucial. It is within this period of time that the majority of adaptive

problems can be identified and, hopefully, rectified. If the foal satisfies all the normal criteria at this age, it has a very good chance of survival.

In a normal birth, the foal is born on its side, lying with its hocks still within its mother and the umbilical cord intact (Fig. 7.12). The newborn foal may be classified as normal, moderately depressed or markedly depressed, according to the scoring systems (Table 16.1; Madigan, 1990; Le Blanc, 1997). The long-term prognosis for the foal is dependent upon this classification.

16.2.1. Anatomical adaptation

Anatomical adaptations can be considered as the milestones that the foal should achieve within set periods of time in order to survive. These parameters can quite easily be observed by the foaling attendant and used to indicate foal viability and identify problems.

Normally the foal should be seen to breathe within 30 s of final delivery. It may take a few sharp intakes of breath as its muzzle first reaches the air during passage through the birth canal, but a rhythm is normally established within 1 min of final delivery. This initial breathing rhythm is normally a steady 60–70 breaths min^{-1} , with a tidal volume (volume of air inspired per breath) of 520 ml, resulting in a minute volume (volume of air inspired min^{-1}) of 35 l (Koterba, 1990). This initiation of breathing results in a

Table 16.1. Scoring system to aid in the classification of newborn foals: score 7–8 normal, score 4–6 moderate depression, score 0–3 markedly depressed (from Madigan, 1990).

Parameter	Score		
	0	1	2
Heart rate	Absent	< 60 min ⁻¹	60 min ⁻¹
Respiration	Absent	Slow, irregular	60 min ⁻¹ , regular
Muscle tone	Limp	Some flexion of extremities	Sternal position
Nasal	No response to stimulation	Grimace, slight rejection on stimulation	Cough or sneeze on stimulation

significant increase in blood oxygen levels. During this first minute of life the foal's heart rate should be in the order of 40–80 beats min⁻¹. This can be measured by placing a hand on the left side of the chest near the heart. The foal's body temperature should be 37.5–38.5°C (Koterba, 1990; Traub-Dargatz, 1993a; Vaala, 1993). The foal's mucous membranes should appear pink and moist and have a refill time of 1–2 s within the first 2 h. Within a few hours, the respiratory rate declines to 35 min⁻¹, with a tidal volume of 550 ml and a minute volume of 20 l (Rossdale and Ricketts, 1980). The heart rate increases on standing to 120–140 beats min⁻¹, plateauing at 80–100 within the first week (Lombard *et al.*, 1984; Lombard, 1990).

The significant increase in blood oxygen concentrations as a result of breathing activates the first reflexes and muscle movements. Within 5 min of birth, the normal foal should be in a sternal recumbency position (Fig. 15.7). It will respond to pain and begin to show evidence of the reflexes associated with rising to its feet, in the form of raising its head, extending its forelimbs, blinking and possibly a whinny (Madigan, 1990; Traub-Dargatz, 1993a). It will also demonstrate the suckling reflex if offered a finger or bottle.

The next major event is the breaking of the umbilical cord, on average 5–9 min after birth. It is considered important that the cord is left to break naturally, as premature breakage is reported to result in the loss of up to 1.5 l of blood by preventing drainage from the placenta (Rossdale and Mahafrey, 1958), though Doarn *et al.* (1985, 1987) failed

to confirm such a blood loss. Breakage naturally occurs at a constriction about 3 cm from the abdomen of the foal. The umbilical artery and vein collapse and constrict naturally at this point, as the pressure of blood circulating within these vessels declines, allowing a clean break, with minimal blood loss and reducing the chance of infection. The mare may be recumbent post-partum for up to 40 min and therefore the cord is usually broken by the movements of the foal in its first attempts to rise to its feet, the increased tension resulting in the cord breaking at the constriction, the weakest point. The umbilical cord therefore breaks naturally after the muscle reflexes of the foal have begun. Once the cord has severed, the navel must be dressed. Traditionally, iodine-based preparations have been used. More recent work suggests that treating with iodine causes a sloughing of skin cells and reopening the navel to infection (O'Grady, 1995). It is suggested that 0.5% chlorhexidine is a better alternative.

Around the time of umbilical cord breakage, the foal makes concerted efforts to raise itself to its feet (Fig. 16.1). The exertion of standing causes the heart rate to increase to up to 150 beats min⁻¹. Heart rate continues to fluctuate with activity around a normal resting heart rate of 70–90 beats min⁻¹ evident in the first few days. When attempting to stand, the series of movements is the same as in the mature horse; stretching forward of the head and neck, extending the forelegs, flexing the hind legs and raising the front end first off



Fig. 16.1. Soon after the end of second-stage labour, the foal makes concerted efforts to rise to its feet; at this time the umbilical cord breaks (photograph Stephen Rufus).

the ground, followed by the hindquarters. Many initial, unsuccessful attempts are made, which form part of the process of developing reflexes and muscle coordination and control. At this stage, the foal is at risk of damage from projecting objects such as buckets, hay racks, automatic water feeders, etc. Successful standing normally occurs in ponies within 35 min post-partum but may take up to half an hour longer in

Thoroughbred foals (Fig. 16.2). Failure to stand within 2 h is indicative of a problem and veterinary assistance should be sought (Rossdale, 1967; Jeffcote, 1972; Madigan, 1990). This remarkable ability to stand and walk so quickly is the result of the evolutionary development evident in plain-dwelling animals, such as the horse, to enable them to flee from potential predators as soon as possible after birth.



Fig. 16.2. Successful standing in the pony foal takes on average 35 min, compared with up to 1 h in the Thoroughbred. At this stage, the foal searches for the udder; the suckling reflex is already present and is elicited by contact with soft, warm, dark areas (photograph Stephen Rufus).

Though the suckling reflex is developed within 5 min post-partum, successful suckling can obviously not occur until after standing and location of and movement towards the udder. The actual reflexes involved in suckling are elicited by contact with soft warm surfaces; hence foals are seen to suckle and nuzzle their dam's flanks while searching for the udder. As soon as the foal stands, it demonstrates directional movement towards the udder, located by the dark and warmth (Fig. 16.2). This process of locating the mare's udder can be easily disrupted by human interference. It is very tempting to try and help a foal and frustrating to watch it suckling at the hock or chest and seemingly unable to locate the teat. However, it is much better to resist the temptation to interfere. The mare will often assist the foal by gently nudging it and moving her hind leg away from her body to allow the foal easier access. Occasionally, primiparous (maiden) mares may need to be held to allow the foal to reach the udder, appearing to be ticklish and initially objecting to the foal's attentions. However, she will soon settle down and should be left alone. In ponies, successful suckling is achieved normally within 65 min; Thoroughbreds are again slower, taking 30 min or so longer (Fig. 16.3; Rossdale, 1967). At suckling, a real affinity develops between mother and foal, which develops into a very strong bond. Human interference may well disrupt this initial bonding process (Chavatte, 1991). The foal, throughout the first few days of life, will suckle at 30–60 min intervals; later on, the interval period lengthens. If during the first few days of life a foal is not seen to suckle for 3 h or so, problems should be suspected.

During the first 12 h the foal should be seen to pass meconium, its first bowel movement. Meconium consists of bowel glandular secretions collected during the foal's intra-uterine life, along with digested amniotic fluid and cell debris, which are passed through the foal's digestive tract *in utero*. Meconium is stored in the colon, caecum and rectum ready for expulsion after birth. Premature expulsion may occur under stressful conditions during or immediately

prior to delivery. Meconium staining of the amniotic fluid or the perineum area of the foal is therefore indicative of fetal or foal stress. Meconium should be brown to greenish brown in colour and is usually all expelled within the first 2 days. Meconium is followed by the characteristically coloured yellow milk dung, which indicates correct gut function (Rossdale, 1967). The routine use of enemas is advocated by some, 12–18 h post-partum (Madigan, 1990). However, their repeated use can irritate the mucosal lining of the gut. Routine enemas are becoming less popular as the adverse effect of such stresses on the newborn foal is increasingly understood. The foal should urinate for the first time within 12 h; colts on average urinate earlier than filly foals (Jeffcote, 1972; Roberts, 1975). Regular urination of small volumes of near-colourless urine should be observed in the normal foal (Rossdale, 1967; Traub-Dargatz, 1993a).

16.2.2. Functional adaptation

This section will consider how the functions of the pulmonary, cardiovascular, temperature-control and immune systems adapt to



Fig. 16.3. In ponies, suckling normally occurs within 65 min, whereas in Thoroughbreds it normally takes 1.5 h.

accommodate the change from the intra- to extra-uterine environment. The change in these functions is reflected in the observed anatomical changes previously discussed.

16.2.2.1. Pulmonary ventilation

Successful gaseous transfer within the lungs across the air–blood interface depends upon their functional and structural maturity. One of the major events is the laying down of surfactant. Surfactant, a complex lipoprotein, provides the alveoli with a surface film, so reducing the surface tension and increasing the efficiency of gaseous exchange and reducing lung collapse (Kullander *et al.*, 1975). Surfactant maturation occurs from day 300 of pregnancy and in some cases not until after delivery (Pattle *et al.*, 1975). Maximum respiratory efficiency is not possible until surfactant development has been completed, and this creates a problem with premature deliveries.

The foal takes its first breath while *in utero* as practice for post-partum functioning of the muscles involved in respiration. Fluid collected within the lungs during pregnancy is expelled by compression of the thorax during delivery and by evaporation and spluttering during early breaths. These first proper extra-uterine breaths and the subsequent breathing rhythm are stimulated by low oxygen (anoxia) and high carbon dioxide plasma concentrations. Cold shock from the atmosphere and tactile stimulus, such as rubbing and mare's licking, all encourage initial breathing. The first breath should occur within 30 s of the foal's hips appearing through the birth canal (Dawes *et al.*, 1972; Gillespie, 1975; Stewart *et al.*, 1984; Ousey *et al.*, 1991). After birth, as a breathing rhythm is established, the alveoli continue to develop, induced by lung expansion and stretching of the bronchi (Vaala, 1993). At birth, the efficiency of gaseous exchange is low – hence the rapid breathing rate. As the bronchi are stretched and alveolar development continues, so increasing the surface area/air ratio, more efficient gaseous transfer is achieved and the breathing rate declines (Gillespie, 1975).

16.2.2.2. Cardiac and circulatory systems

In utero, the placenta acts as the 'lungs' in being the major site of oxygen and carbon dioxide exchange, as well as nutrient uptake. In order that the placenta is supplied, blood must pass from the pulmonary artery via the ductus arteriosus to the aorta, so bypassing the pulmonary system (the lungs) and passing directly to the placenta (Fig. 16.4). Only a small supply of blood to the pulmonary system is required, adequate for pulmonary growth and development. Additionally, the foramen ovale allows blood to pass from the right atrium directly to the left atrium and hence immediately around the body via the aorta, rather than to the right ventricle. Blood enters the placenta via the two umbilical arteries and leaves via the single umbilical vein, to pass to the liver and then back to the right side of the heart. The bypassing of the lungs is aided by the relatively high pulmonary vascular resistance compared with the systemic resistance. Both ventricles work in parallel, with the right ventricle dominating in size and output (Vaala, 1993).

Immediately post-partum, the circulatory system of the foal must change dramatically to redirect blood through the pulmonary system to the lungs, and away from the umbilical system to the placenta (Fig. 16.5). The trigger for this change is unclear, but a decrease in pulmonary resistance plays a role. As the foal takes its first few breaths, the collapsed lungs inflate, stretching the alveoli and rapidly reducing pulmonary resistance, resulting in increasing blood perfusion of the lungs (Kullander *et al.*, 1975). As pulmonary resistance declines, blood is drawn up directly through the pulmonary vein to the lungs and not across the ductus arteriosus to the aorta. As more blood enters the left side of the heart from the pulmonary artery, blood pressure increases. When blood pressure in the left-side of the heart is greater than that in the right, the foramen ovale closes. Some blood leaving the left-hand side of the heart to the aorta may still continue to pass through the ductus arteriosus to the pulmonary system to recirculate through the lungs. This continues until the closure of the ductus arteriosus at 3–4 days of age

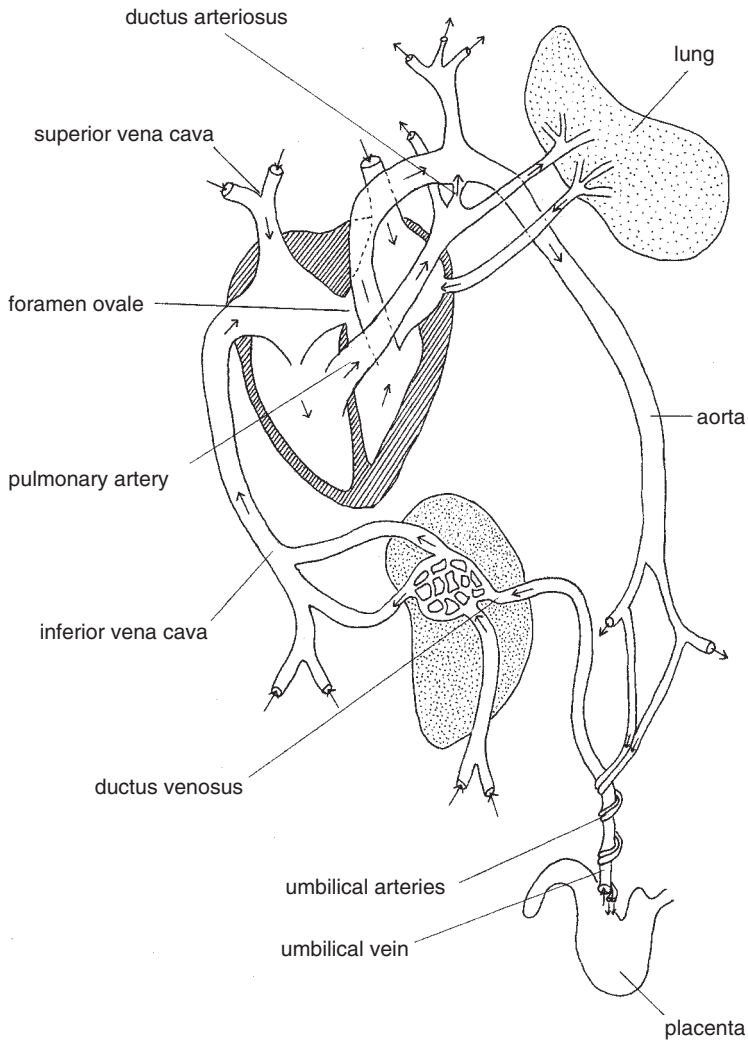


Fig. 16.4. The fetal circulatory system *in utero*. The blood passes through two openings, the foramen ovale in the atrium wall and the ductus arteriosus, hence bypassing the lungs.

(Rossdale, 1967; Button, 1987; Lombard, 1990). Closure of the ductus arteriosus is thought to be associated with increasing plasma oxygen concentrations and decreasing tissue concentrations of prostaglandins (Vaala, 1993). Until its complete closure, it may reopen in response to stress or hypoxaemia (Cottrill *et al.*, 1987). Blood is now pumped from the right atrium via the pulmonary artery to the lungs for oxygenation and back via the pulmonary vein to the heart for circulation around the body (Figs 16.4 and 16.5).

Many newborn foals initially suffer from arrhythmia – irregular heartbeat – but this soon settles down naturally. Many foals show signs of asphyxia during the second stage of labour, evident as a blue tongue and mucous membranes of the eyes, caused by a reduction in blood flow and therefore oxygen to the head. Such constriction of the head, neck and chest during passage through the pelvis is of no long-term significance, providing the foal continues to be delivered normally and parturition is not delayed. After birth, the

Fig. 16.5. The circulatory system of a normal foal post-partum. The foramen ovale and the ductus arteriosus close within a few days of birth.

mucous membranes may remain blue/grey in colour for a short while, but should be the normal pink colour within 2 h.

At birth, the red-blood cell count is considerably elevated compared with that later on in life. This is thought to be due to fetal stress during birth, as levels are further elevated in foals born with difficulty. Within 2 h of birth, red blood-cell counts decline and white blood-cell counts rise to normal levels (Chavatte *et al.*, 1991).

16.2.2.3. Temperature

The foal is born with a well-developed temperature-control mechanism, unlike many

other mammals, especially humans, which cannot effectively control their body temperature for several weeks after birth. At birth, the foal can maintain a steady body temperature of 37–37.5°C (100°F) (Rossdale, 1968), which increases to 38–38.5°C within 1 h, despite a cold environment. This is due to the high metabolic rate of newborn foals (200 W m⁻²), which is three times that of a 2-day-old foal (Ousey, 1997). The exact mechanism by which it maintains this steady body temperature is unclear but is likely to involve the ability to shiver, which is first evident in newborn foals during the first 3 h post-partum. In addition, heat produced by muscular activity and the strain of the foal's first move-

ments is likely to contribute to maintaining body temperature, along with the foal's insulating layers of fat and its hair coat. Unlike the human baby, the foal does not have brown adipose tissue. Its ability to shiver earlier in life negates the requirement for brown fat, which is a heat-producing tissue. The presence of brown fat is associated with neonates that are unable to shiver and have less fine control over their body temperature (Ousey *et al.*, 1991). The onset of hypothermia in newborn foals may be rapid and can result from infection, as well as dystocia and a cold environment. Hypothermia may cause hypoxaemia and acidosis, causing an attempted reversion to fetal circulating patterns and altered gastrointestinal function.

16.2.2.4. Immune status

As discussed in Chapter 5, the equine placenta is epitheliochorial and, as such, presents a considerable barrier to the passage of blood components from mother to fetus *in utero*, especially those of large molecular size, such as immunoglobulins (antibodies). In the foal, therefore, the attainment of immunoglobulins *in utero* is limited and so colostrum is vitally important for achieving adequate immunity for survival in the extra-uterine environment. At birth, the foal is plunged from sterile conditions into an environment of varying immunological challenge. The foal's system is perfectly capable of meeting this challenge by producing its own antibodies over time, but is born immunologically naïve (without antibodies), apart from a small concentration of immunoglobulin M (IgM). The foal requires immunological protection until it has produced enough antibodies to protect itself. The biggest contributor to passive immunity is colostrum. Equine immunoglobulins can be subdivided into IgG, IgM and IgA. The most predominant in colostrum is IgG and it is this that is most evident in the circulation of the young foal (McGuire and Crawford, 1973). Adult levels of immunoglobulins are not immediately evident in the foal; these are reached only after the foal starts to actively produce its own immunoglobulins. For the

first 24 h post-partum, enterocyte cells lining the foal's small intestine are able to absorb by pinocytosis large protein molecules such as immunoglobulins (Jeffcote, 1987). The ability to absorb whole proteins is seemingly enhanced by other components of colostrum and controlled in part by cortisol, though the exact mechanisms are unclear. Over time, the enterocytes are replaced by cells incapable of absorbing proteins. It is therefore essential that newborn foals receive colostrum, 500 ml at least, within the first 24 h of life and preferably within the first 12 h, when absorption is most efficient (Pearson *et al.*, 1984; Jeffcote, 1987). This ensures that the foal obtains maximum protection from infection via maternal antibodies. Several tests to ascertain the immunological (IgG) status of the foal are available (Bertone and Jones, 1988; Le Blanc, 1990; Madigan, 1990). It is generally agreed that foal IgG serum concentrations of 400–800 mg dl⁻¹ are appropriate. Colostrum may also be tested and it has been suggested that a specific gravity of > 1060 is indicative of an IgG concentration of > 3000 mg dl⁻¹ which, in turn, will result in a foal serum concentration of > 500 mg dl⁻¹, adequate for foal protection (Le Blanc *et al.*, 1986).

The foal's own immune system does start to function to a very limited extent during pregnancy; hence a small concentration of IgM is evident at birth, but it does not reach maximum capacity until 3–4 months of age. Colostrum therefore provides protection in the interim until the foal's own immune system is fully functioning (Rouse and Ingram, 1970). If the mare is immunized during late pregnancy, then the antibodies raised pass to her colostrum and are available to the foal, providing it with essential temporary protection. Such immunization is advised 4–6 weeks prior to expected delivery.

16.2.3. Biochemical adaptation

The foal's metabolic system at birth undergoes dramatic alterations from a dependent to an independent status. While *in utero*, it is entirely dependent upon the maternal system via the placenta; post-partum this

dependency is removed and replaced by reliance upon the pulmonary and gastrointestinal systems.

At birth, the foal goes through a transitional period after the severing of the maternal connection and before suckling. This period of time is one of considerable stress and exertion, for which energy is required, provided by liver glycogen stores laid down during the later stages of gestation. The equine fetus does not store glycogen within the brain. Glycogen is mobilized in the form of glucose immediately post-partum and supplies, in an easily convertible form, the energy required for this transition period. Glucose levels can be measured in the plasma of newborn foals and used to indicate the availability of these glycogen stores. However, these stores are finite and are quickly depleted in cases of stress/hypoxaemia, etc. Immediately post-partum, glucose concentrations should be in the order of 50–70 mg 100 ml⁻¹ blood. Levels lower than 50 mg 100 ml⁻¹ indicate hypoglycaemia. Once the foal has suckled, glucose levels increase and in a normal 36 h old foal reach values of 100–110 mg 100 ml⁻¹ blood.

Bicarbonate levels rise steadily over the first 36 h of life from the 23–28 mmol l⁻¹ evident at birth (Jones and Rolph, 1985; Fowden *et al.*, 1991). Lactate concentrations also rise immediately post-partum and decline to normal adult levels within 36 h. The initial increase in lactate coincides with a fall in venous pH (7.4 post-partum to 7.35 at 30 min) and this may be a result of the energy demands during the transition period (Stewart *et al.*, 1984). This fall in pH rectifies itself within 12 h. Plasma cortisol levels also rise significantly immediately post-partum. The significance of this is unclear, but it has been postulated in some mammals to be associated with the final maturation of internal organs, such as the respiratory system.

The biochemical changes apparent in the newborn and very young foal can only be assessed via blood sampling and they vary considerably. They must therefore be viewed with a certain amount of caution when used as a diagnostic aid (Jones and Rolph, 1985; Fowden *et al.*, 1991).

16.2.4. Post-partum foal examination

Within 1 h of birth, it should be evident whether or not the foal is adapting to the extra-uterine environment appropriately. If problems are suspected a veterinary surgeon should be called. Assessment of the following parameters, some of which have already been detailed, will give a very good indication of foal well-being.

Foal

- Heart rate
- Respiration
- Ability to stand
- Vigour
- Ability to suckle
- Straight legs
- Body weight
- General demeanour

Placenta

- Weight
- Integrity
- Abnormalities

Mammary-gland function

- Colostrum quantity and quality

16.3. Early Foal Management – the First 6 Weeks

Management of the lactating mare and foal depends to a certain extent on whether she is to be returned to the stallion or not. If the mare is to foal at stud, her early management and that of the foal will largely be determined by the general stud management and practice. As such management has a significant effect on the foal's long-term prospects, this is an area that should be discussed during initial stud selection.

Immediately after foaling and foal examination, the following management procedures may be carried out:

- Navel dressing
- Administration of broad-spectrum antibiotics (penicillin/streptomycin) – a precautionary measure taken by some
- Administration of vitamin supplements
- Blood testing

Blood-cell count

Isoerythrolysis test – mare–foal compatibility test

Immune-status test/immunoglobulin uptake

Enema – still practised routinely by some, but not advised unless meconium retention is suspected

Apart from this, the mare and foal should be left in peace, with regular, unobtrusive observation. A radiant-heat lamp may be used to provide warmth for the foal. As indicated, antibiotics may be given as a precautionary measure; however, indiscriminate use is not encouraged, due to the potential for bacterial resistance, disruption of the colonization of the small intestine and caecum, diarrhoea and selection for antibiotic-resistant *Salmonella* organisms (Madigan, 1990). Enemas are routinely administered by some. If used, they are best administered during the first passage of meconium or at 12 h. Administration too early may have no effect if no meconium has yet passed beyond the pelvic inlet (Madigan, 1990). No controlled trials appear to have been carried out on the use of enemas and many now discourage their routine use.

16.3.1. Exercise

For the first 3 days of life, the eyesight of the foal is not accurate enough to safely allow it out of the stable or small foaling paddock. After 3 days, it should have developed adequate appreciation of distance and depth to be turned out with its dam for an hour or two during the day, providing the weather is good (Fig. 16.6).

It is best to avoid turning out young foals if it is wet or very cold and windy. Such weather will easily soak the foal through and, as both mare and foal will be reluctant to move around in such weather, it defeats one of the main objectives of turning them out, that of exercise (Back *et al.*, 1999). The paddock provided for foals should be small; half an acre is ideal. It should have strong, well-constructed fences, ideally post and three rails with no wire. There should be no protruding objects, old machinery, wire, holes, low branches, etc. as these can prove death traps for young foals still not that steady on their feet. An alternative system used in Europe, especially France, is electric-fence paddocks. Providing mares are accustomed to electric fences, large fields can be



Fig. 16.6. After 3 days, a foal's eyesight should have developed enough to allow it to be turned out with its mother (Penpontbren Welsh Cob Stud).

divided up into small paddocks with electric tape, allowing association with other mares and foals but security in the first weeks (Fig. 16.7).

Water should be provided in a bucket. Streams or large water troughs can also prove lethal for a very young foal. The paddock should have plenty of good grass, as this will encourage the mare to eat, which is especially important in mares whose appetite has declined after parturition.

Persuading the foal to leave its stable for the first time can be a challenge, but should be made as free from trauma as possible; otherwise nervousness will be perpetuated. During the first 3 days of life the foal should have been handled gently, stroked all over and got used to having arms put around it. When the big day comes, therefore, it should be used to human contact. The best day to turn a foal out for the first time is a nice sunny day, but not too hot or flies will be a problem, and bright sunlight may discourage the foal from leaving the much darker stable environment. There should be at least two handlers. The mare should be led ahead slowly by one handler and another should cradle the foal in his/her arms, one arm behind its hindquarters and the other around its chest, and encourage it to follow its dam (Fig. 16.8).

Some foals will follow easily; others prove more difficult. A foal should never be pulled from the head by means of a halter, as this may seriously damage its neck and head. A soft twisted cloth, bandage or thick rope can be put around its neck initially, which can later be replaced with a soft leather or webbing halter. Leather is preferred, as it will stretch and eventually break under strain. Some people like to leave head collars on foals while they are out; this can be very convenient for catching them and gives them time to get used to them. However, the collar must be very well fitting to ensure that it will not get caught on anything or allow the foal to catch its feet in it (Fig. 16.9).

16.3.2. Handling

Initial handling in the first few days before turnout should consist of gentle stroking over the whole body and general familiarization with humans. Once a foal can be led, it must start to learn how to be tied. This is best done by using a round pole with no projections, so that the foal cannot get itself twisted up or caught on fences. A rope can then be attached to its head collar and on to the pole. As mentioned earlier, there is a risk of damage if the



Fig. 16.7. Large fields can be divided up into small, single mare-and-foal paddocks using electric fence tape, providing contact with other mares and foals, along with security from them during early life.



Fig. 16.8. The foal may be encouraged to lead for the first few times by cradling the foal in your arms, one arm behind its hindquarters and the other around its chest (photograph Angela Stanfield).



Fig. 16.9. A leather halter is preferred as a first-time halter, as it will stretch and eventually break under strain (photograph Pauline Norris).

foal is pulled by a rope attached to its head. Hence an alternative is to loop the rope around the foal's girth and up through its head collar and on to the pole. This method of restraint means that all the pull is taken on the girth and not the foal's head. However, foals will soon learn that they cannot escape and that it is easier to stand still.

Once the foal has learnt to accept tying, the general stroking and handling can progress to grooming and attention to feet

and eventually travelling. These are particularly important if you intend to show the foal. Grooming can develop slowly and the foal will soon come to enjoy it, providing all progression is done slowly and patiently (Figs 16.10, 16.11 and 16.12).

Providing the weather is good, a foal can be bathed – again, of great use if it is to be shown. The weather must be warm and it should not be bathed very early or late in the day, to avoid it catching a chill.



Fig. 16.10. A foal should learn to be tied as one of its first lessons.



Fig. 16.11. Once the foal has learnt to be tied, it can be groomed.

Introduction to a trailer or lorry can also be done in the first 6 weeks of life. The mare can be used to encourage the foal and many take to it easily, providing the mother is a good loader. If she is not, there is a danger of the foal picking up her bad habits or her fear. In such cases, leaving the trailer in the foal's paddock with the door open and feed in the top end can encourage it to investigate and get used to going in and out at will. If this is done, the foal must be watched at all times to ensure that it does not hurt itself. Once the mare and foal have been successfully loaded and unloaded a couple of times, they can be

taken for a short ride. A foal will sometimes travel better if there is no central partition dividing the trailer. If there is a top door, it should be closed or a bar or cover used to prevent the foal from trying to escape over the tailboard if it panics.

16.3.3. Feet care

The foal's feet should need little attention in early life unless they have a significant deformity. Nevertheless, picking up the feet, picking out the hooves and grooming the



Fig. 16.12. The foal should also learn to have its feet picked up, though they should need little attention at this stage.

legs should be done regularly, and these, along with ensuring a general acquaintance with the blacksmith when the mare's feet are attended to, will ease work on foal's feet later on. Regular inspection of the feet will allow examination for injury and damage.

16.3.4. Behaviour

During the first 6 weeks of life, the foal shows quite significant development in behaviour and social interaction. Initially, the foal's whole world and social experience revolves around only its mother. This includes play, which may consist of rubbing her mane and tail and kicking. Through this, it begins to learn how far it can push it before being reprimanded and so what is acceptable and what is not. Once the foal has developed more steadiness on its feet, normally after about a week, it will start to explore further away from its mother, but never straying far. Over the next few weeks, the circle gets bigger and it spends more time away from its mother, investigating and playing alone (Fig. 16.13).

If at this stage the foal has access to other foals, it will begin to interact with them and play will gradually include them rather than its mother. By 8 weeks, it spends up to 50% of its time playing with other foals and only 10%

playing around its mother. If, however, the foal has no contact with others, it will play with its mother much longer and may try to play with other older horses present or even dogs or other animals regularly in its company. If its mother is particularly possessive or shy, these characteristics can be passed on to the foal and it will not integrate as well with other foals (Carson and Wood-Gush, 1983b).

Apart from play, the foal spends a significant amount of time lying down resting (Fig. 16.14). These are normally short periods of rest, particularly in warm sunlight, between periods of play.

The remainder of its time is spent suckling. These periods of suckling in the first week are short and may occur as often as every 15 min. With time, the intervals between sucklings become longer – 35 times day⁻¹ by week 10 – but the periods of time spent suckling and hence the intake increase (Carson and Wood-Gush, 1983a,b; Fig. 16.15).

16.3.5. Introduction to solids

The foal may be first seen investigating concentrate feeds, and even ingesting some, as early as 3 days of age. Investigation of the mare's feed at an early age is to be encouraged, as the mare's milk is naturally short of iron and copper, which invariably causes



Fig. 16.13. The foal's circle of play becomes increasingly larger over the first few weeks of life, as it gains independence (Penpontbren Welsh Cob Stud).



Fig. 16.14. Apart from play and suckling, the foal spends a large proportion of its time lying down and resting.

anaemia in very young foals. Copper and iron are vitally associated with red blood-cell function and haemoglobin levels. Adequate levels of copper and iron can be achieved by the foal picking at the mare's feed. Anaemia may persist in foals too weak to nibble hay or concentrates until they are treated or are able to eat (Fig. 16.16). Foals may also be observed nibbling the mare's dung in the first 5 weeks or so. The reason for this is unclear but may be a means of addressing mineral deficiency or introducing bacteria

into the gut in order to set up the appropriate microflora for forage digestion. This coprophagic behaviour does, however, run the risk of parasite ingestion by the foal and so worming of mares with young foals at foot is particularly important.

In theory, the mare's milk up until peak lactation, week 6–8, provides the vast majority of the nutrients required by the developing foal. During this time, however, gradual investigation and ingestion of its dam's feed provides an increasing amount of nutrients,



Fig. 16.15. As the foal grows up, it spends less time suckling its mother, the intervals between suckling getting larger (Penpontbren Welsh Cob Stud).



Fig. 16.16. A foal will soon be seen investigating its mother's feed. This is to be encouraged, as it provides the foal with essential copper and iron.

but this is not significant until after 6–8 weeks. The amount of extra creep feed that a foal will require in the first few weeks depends largely on the mare's milk yield. In

addition to concentrates, the foal must be introduced to roughage in the form of grass or hay, as a diet of concentrates and milk alone can cause diarrhoea.

Creep feed can be introduced as an optional extra as early as 1 week, but the foal should never be forced to eat it. The progression from milk to solid food must be gradual and can start slowly at an early age. Many proprietary creep feeds are available on the market, specially formulated to ensure that the foal gets adequate nutrients for a healthy start in life. If you mix your own, there are a few considerations to bear in mind. Protein should be relatively high in foal diets when compared with adult diets. In particular, these proteins should be digestible and contain the ten essential amino acids for horses: lysine, methionine, leucine, isoleucine, histidine, arginine, tryptophan, valine, phenylalanine and threonine. Many legumes, grains and pulses tend to lack lysine, and soybean meal and linseed meal can be added, within reason, as good suppliers of lysine. Other dietary components of special importance in growing animals are calcium and phosphorus. A ratio of these two minerals of 2:1 should be aimed for. These minerals are essential for healthy growth of bones, cartilage, tendons and joints. Excess calcium or phosphorus can cause problems. Excess phosphorus causes calcium to be mobilized

from the foal's bones in order to maintain the ideal 2:1 ratio, causing bone weaknesses and epiphysitis and so delaying growth. Many people supplement calcium in the form of limestone flour or equivalent, as many legumes, grains and pulses have a relatively high concentration of phosphorus. Low copper concentrations are reported to be associated with angular limb deformities and high zinc concentrations with osteochondritis dissecans.

In addition to concentrates, foals should have access to fresh green forage. Lucerne is good, as it is relatively high in digestible protein and calcium. Hay may be fed, but it must be of a good quality, with no evidence of dust, mould, dampness, etc. Best of all is free access to fresh grass, which provides an ad lib supply of continually fresh material.

Once the foal starts to pick at its mother's food, care must be taken that, when its intake becomes significant, she receives enough to meet her requirements. Foal and mare should now be fed individually. By 3–4 months, the foal should be eating 1 kg day^{-1} – about 0.25–1% of body weight. Careful monitoring of the foal's feed is required to prevent obesity and resultant conditions, such as developmental orthopaedic disease (Ralston, 1997; Coleman *et al.*, 1999).

16.3.6. Dentition

Providing the teeth of the foal erupt as expected and at the correct angle, there is no need to do anything with the teeth in the first 6 weeks. Most foals are born with the central incisors or they erupt within 8–9 days; the middle incisors should then erupt at 4–6 weeks (Table 16.6).

16.3.7. Immunization and parasite control

Foals are often routinely immunized against tetanus in the first few days of life. However, colostrum from a suitably immunized mare is a much more effective method of providing protection against tetanus and numerous other diseases. Recent work suggests that

immunization of very young foals, born to mares that are adequately protected by vaccination, may have a detrimental effect on their long-term protective response to subsequent immunization (Maanne *et al.*, 1992). In some countries, it may be worth considering immunization against strangles or rabies, depending on the prevalence of the disease. Worming can be first done at 7 days of age, the foal should then follow a regular worming regime, worming against ascarids and *Strongyloides westeri* in both the mare and the young foal is particularly important (Clayton, 1978; Craig *et al.*, 1993; Shideler, 1993d).

16.4. The Orphan Foal and Foals Requiring Additional Support

The inability of a dam to bring up her foal due to her death, illness or injury can be a serious setback for the foal. The newborn foal depends on its mother for a variety of things, including colostrum, nourishment via milk, maternal affection and psychological development. Any replacement dam has to satisfy all these needs. Survival rates in orphan foals were at one stage very low. However, research and development have now allowed the needs of the foal when changing from newborn to weanling to be better understood and the management of the orphan foal geared to suit these needs.

The treatment required by the foal depends upon when the problem occurred, under what conditions and the reason for additional support. For example, foals that have received no colostrum or a very limited amount need to be identified and fed colostrum immediately. If the mare died during parturition colostrum may be milked from her and fed to the foal. Foals that have been orphaned after 48 h or so may have been able to suckle and should have received enough colostrum for adequate protection. Colostrum may be collected and frozen for up to 5 years, though it is suggested that some degeneration starts to occur after 12 months. Some studs routinely collect and freeze extra colostrum from mares that produce excess or from those that lose their

foals. Colostrum substitutes are available; colostrum from other animals was traditionally used but is now frowned upon, due to digestive upsets arising from differences in composition. Alternatively, serum transfusions may be given to provide the foal with immediate protection.

In addition to classifying a foal according to its colostrum intake, its state of health and strength should also be assessed. Thus any assessment should compare the parameters of the orphan foal against those expected of a normal foal. A difficult birth due to dystocia, in addition to the death of the dam, will result in an orphaned foal requiring immediate intensive care, such as stomach tubing with colostrum; antibiotic treatment, precautionary against infection; multivitamin injection; oxygen replenishment; or even blood transfusion. On the other hand, a strong orphan may readily suck from a bottle and not require additional treatment.

Apart from death, a dam may for some reason be unable to raise her foal herself. The reasons for such failure are important when deciding upon the appropriate management. A mare may be unable to feed her foal through inadequate milk production, due to mastitis or a physical abnormality. In the case of mastitis, the inability to produce milk may be permanent or temporary. If temporary, the foal may only need supplementary feed to keep it going until the infection has cleared. If a mare seems physically unable to produce enough milk, her foal may be supplemented to a certain extent but left hungry enough to encourage it to suckle its mother. This suckling may then stimulate her mammary glands to increase their milk production. Occasionally, foals may need supplementing just for the first few days of life, especially if they are premature, so giving the mammary gland time to catch up and secrete adequate milk. In such cases, the foal need not be removed from its mother, allowing the mother-foal bonding to continue: psychological damage to the foal is thereby limited.

Occasionally, if the birth has been particularly stressful or in the case of some primiparous mares, the foal may be rejected by its mother. This rejection may vary from disin-

terest to physical attack. Such mares may be sedated temporarily or the procedures used for fostering foals, discussed later, can be applied. Rejection is often only temporary, the mare subsequently accepting the foal. During such incidences of weak mare-foal bonding, human interference must be minimized, as any external stimulus will only serve to worsen the situation. Intervention must be used only if the foal's well-being is at risk.

The vast majority of orphan or bottle-fed foals are at a disadvantage as far as health status is concerned. Health care for such foals is therefore very important. All the routine parameters for the newborn foal should be checked at birth. It is also advisable to check temperature and heart rate twice daily for the first few weeks to allow rapid identification of problems, so that immediate treatment or further investigation can commence. The foal may be helped by multivitamin and antibiotic injections.

Orphan foals and those requiring additional support are more susceptible to the ordinary infections that normal foals take in their stride. They should be watched carefully and kept in a scrupulously clean environment. Diarrhoea is a common complaint of bottle-fed foals, especially if fed artificial diets. The diarrhoea may be a direct result of the diet but may also be a sign of infection. Persistence after 24 h should be considered as serious, as dehydration can bring a foal down very quickly. Respiratory diseases can also be a problem in orphan foals, so their housing should be draught- and damp-free, while providing good ventilation. Meconium retention may also be a problem, especially in foals that have not received adequate colostrum, as colostrum acts as a laxative. An enema may be required and the foal watched for subsequent constipation.

As discussed, the loss of a mare deprives the foal of nutritional and psychological security. A foal can survive without the psychological stimulus of a mother, though this may affect its long-term behaviour. It is not, however, able to survive without nutrition. There are several ways of providing that nutrition.

16.4.1. Fostering

The fostering of a foal on to a mare that has lost her own foal is the best solution. This provides the foal with a source of nutrients and psychological security, and provides the mare with a substitute for the lost foal. The mare used should be as close in her stage of lactation as possible to the foal's natural mother. This is ideal as the nutritional components of the mare's milk vary with time of lactation and are coordinated with the foal's developing requirements (Chapter 9).

If no such foster-mother is available, but there is a mare that has lost her foal in the last few weeks, then she can be used as a companion to the foal and will be an invaluable support to its psychological development. In such a case, the foal may be supplied with artificial foal milk, designed for the very young foal, in addition to being allowed to suckle its foster-mother. If the foal shows signs of poor health, it can be prevented from suckling its foster-mother and fed specially formulated foal diet for its age, but it will still have her psychological support.

Within the UK and in other countries 'foaling banks' exist, which arrange the teaming up of orphan foals and mares. Multiple suckling – the introduction of an orphan foal to a mare that has plenty of milk but with her own foal still at foot – is not very successful in horses. This technique is successfully practised with cattle, where milk yields are artificially high. In horses, multiple suckling tends to lead to two poor foals and may lead to resentment between them. In North America, nurse mares are available as foster-mothers. These mares are exceptionally good milkers and are kept primarily for leasing out as foster-mothers, after their own foals have been weaned early.

Once a foster-mother has been found, it can be quite tricky persuading her to accept the orphan foal. She is much more likely to accept a foreigner if it smells of her or her own foal. Tricks such as rubbing the placenta of the foster-mother over the foal or skinning the dead foal and placing the skin over the orphan work quite well, but depend upon the handler, the mare and the foal being all

together at the right time. Later on, the orphan foal can be rubbed with the mare's urine, faeces or milk, especially in the head, neck, back and navel region, again to mask its own foreign smell. Other tricks are used, such as rubbing strong-smelling substances on the mare's nose and over the foal in an attempt to disguise its smell.

Once prepared, the foal should be introduced to the mare with great care. One person must hold the mare and at least one hold the foal. They should be introduced to each other at the mare's head end and her reaction noted very carefully. If all goes well and she shows no objection, the foal can be allowed to slowly explore around the mare, making sure that plenty of assistance is available if the mare should object. The foal can then be introduced to the mare's udder and allowed to suckle, providing again the mare shows no signs of objection. Rarely will things go according to plan and the mare will often initially show signs of annoyance or objection to the orphan, in which case it should be removed and reintroduced to her head end again. Eventually, most mares will let the foal suckle and, once this has occurred, it can be left in a stable with her unrestrained for a period of time. The area should be relatively confined so that they remain in close contact with each other and the foal does not become isolated. Throughout this period, observation at all times is very important and immediate action taken to remove the foal if the mare starts to object. Slow, patient progress will pay off and, once the foal has suckled several times, the mare will rarely object to it. After a couple of days, they can be turned out into a small paddock alone to help develop the bond between them over a distance before introducing them to the *mêlée* of other mares and foals.

Problems with fostering do occur. Some mares, regardless of all persuasion, will not accept a foal and it should be given up as a bad job before humans and horses lose patience and the foal experiences yet another rejection. There are, however, various techniques to help persuade reluctant foster-mothers, including the use of a crate in

which the mare is held such that she is unable to turn around. The sides of the crate should be solid with a hole at the end nearest the udder through which the foal, which is outside the crate, can suckle. This can work quite well and after the foal has suckled several times the mare can be removed from the crate and she will often accept the foal. Other tricks are used to elicit the mare's protective response: the introduction of a dog or another mare within sight can induce a protective response in the mare towards the foal, which often leads to acceptance, but the applicability of such means depends on the individuals concerned.

If all else fails, a nanny-goat may be used as a foster-mother. It can be placed on an elevated platform for suckling, and its continual presence provides company for the foal. However, care must be taken, as goat's milk is not of the same composition as horse's milk, though it is nearer than cow's. It has two-thirds the sugar content and three times the fat content of mare's milk and tends to cause gastrointestinal upsets, especially associated with gas retention (Chapter 9).

16.4.2. Artificial diets

If it is not possible to find a foster-mother or if a foal requires supplementary feeding, there are specifically formulated equine milk substitutes available on the market (Table 16.2). These are formulated to mimic the mare's natural milk components (Frape, 1998). All these dry powders must be mixed with water under sterile conditions and fed at 37.5°C. Other formulas are used, based on cow's milk or dried cow's milk, with added components to make up the shortfalls in cow's milk compared with mare's milk. These are not very successful, though still popular, and often result in gastrointestinal upsets, causing diarrhoea, dehydration and, if not rectified, a rapid decline in foal development and growth. If diarrhoea does occur, then milk substitutes should be replaced by a 50% glucose–electrolyte solution, made up in sterile water, for 1–2 days, with the slow reintroduction of the milk substitute. Ensuring

that regular small feeds are fed rather than fewer large ones can reduce the incidence of diarrhoea. A young foal will naturally suckle seven times h^{-1} (Chavatte, 1991). It is therefore necessary that frequent feeding is gradually reduced from once every 1–2 h during the first few days to every 2 h after 2 weeks of age (Carson and Wood-Gush, 1983b; Naylor and Bell, 1987). Specially designed mare's-milk substitutes are widely available nowadays and there is no excuse for feeding other formulas except in real emergencies (Green, 1993; Frape, 1998).

During the first few days of life, the orphan foal should also be given a broad-spectrum antibiotic plus a vitamin injection, to give it an extra boost over what is to the foal a very stressful time. The foal should be watched carefully and help called at any signs of trouble. Orphan foals tend to go downhill much more quickly than those with their mothers. For the first few weeks, the orphan foal will need to receive its milk via a bottle, and a large plastic squash bottle

Table 16.2. An example composition of a foal milk replacer for use with orphan foals or those that require additional nutritional support* (from Frape, 1998).

Component	%
Glucose	20.0
Fat-filled powder (20% fat)	5.0
Spray-dried skimmed milk powder	40.0
Spray-dried whey powder	32.7
High-grade fat†	1.0
Dicalcium phosphate	1.0
Sodium chloride	0.2
Vitamins/trace elements‡	0.1

*Disperse in clean water at a rate of 175 g l^{-1} . May also be pelleted and mixed with a stud mix as a weaning feed for orphan foals.

†High-quality tallow and lard, including dispersing agent. Stabilized vegetable oil could be alternatively added at the time of mixing.

‡To provide vitamins A, D₃, E and K₃, riboflavin, thiamine, nicotinic acid, pantothenic acid, folic acid, cyanocobalamin, iron, copper, manganese, zinc, iodine and selenium.

with a lamb teat works quite well. It is very difficult to prescribe how much and how often it should be fed, as there is no hard and fast rule. However, as a rough guide, 110–220 g of milk should be fed every hour in the first week to ensure a daily intake of 9–10 MJ digestible energy (DE). Over the first few weeks, daily intake should increase to 9–18 l, and with time the frequency of feeding can decrease (week 2 every 2 h, week 3 every 3 h, week 4 every 4 h). After 4 weeks, it may be fed just four times day⁻¹. The exact timing and amount depends on the introduction and acceptance of milk in a bucket and/or concentrate feed. After about day 3–5, made-up milk powder can be placed in a bucket and hung at a level giving easy access for the foal. This will allow it to wean itself slowly off the bottle and on to the bucket. Some foals feed from the bucket very quickly; others need more encouragement. It must be ensured that the milk is always clean and fresh; otherwise the foal will be discouraged from taking it. Warm milk and encouragement by licking off human fingers can help a reluctant foal to accept bucket milk (Green, 1993; Frape, 1998).

Some foals, instead of moving over to bucket feeding, are kept on bottle feeds by use of an automatic feeder. The more sophisticated automatic feeders mix and regulate the amount and temperature of the milk delivered, and the foal sucks through a conveniently placed teat in its pen wall. Less sophisticated machines require manual refilling with pre-mixed milk. Either, if accepted, reduces the labour, especially at night, if there are several orphan foals to be fed. It is very important that such feeders are regularly stripped down and sterilized, as bacteria build up rapidly in such an ideal environment and can cause the foal immense problems.

16.4.3. Introduction of solids

Many breeders recommend the introduction of solids as early as 1 week of age, though the foal's intake at this time will be very limited. Introduction at an early age gives the foal time to become accustomed to solid feed and

gradually wean itself off milk. This progression from milk to solids can be helped by introducing foal milk pellets as a halfway house. As soon as the foal is eating a regular amount of concentrates, then the milk pellets fed can be reduced. Ad lib access to fresh green grass, or alfalfa as a substitute, will also encourage a foal to eat. By 2 weeks of age, the foal should be turned out, at least for part of the day, with a companion, weather permitting, into a small safe paddock. This will introduce it to fresh grass and allow it room to stretch its legs, experiment with movement and investigate its new environment.

By 2 months of age, most orphan foals are consuming enough concentrates to allow them to be weaned off milk. The exact time will depend on the foal's well-being, and final weaning should not be attempted unless the foal is fit and healthy, as it will always result in a slight setback. Orphan foals can be fed normal foal creep feeds as discussed earlier (see section 16.3.5).

By 3 months, the foal should be completely weaned off milk and fed on a ration of concentrate to supplement fresh grass and forage. With orphan foals in particular, a close watch should be kept on the foal's body weight and its diet should be changed to accommodate any significant rise or fall in condition. As with normal foals under- or overcondition can be detrimental to growth and development (Ralston, 1997; Coleman *et al.*, 1999).

16.4.4. Discipline and the orphan foal

One of the problems encountered more often with orphan foals than with those brought up by their dams is bad behaviour. The presence of the mare acts to discipline the foal, teaching it respect for others, both horses and humans, and passing on, it is hoped, good behavioural characteristics. Hand-reared foals can become overfriendly with and lack respect for human beings, treating them much as they would other foals and horses, with nipping, kicking, chasing, etc. If this is allowed to get out of hand, such animals can be extremely hard to handle in later life.

To avoid this, orphan foals should be allowed sight of other horses as soon as possible. Direct contact may be dangerous at such a young age but sight and smell will help. By the time they are 1–2 months old, they should have an established gentle companion – one that is placid and does not bully the foal; Shetland ponies and/or donkeys are often used. By 4–5 months old, they should be able to cope with other weanlings and can be run out together, allowing play and social interaction with their fellows. Discipline while in the company of humans should continue to be strict and consistent.

16.5. Mare Management in Early Lactation – the First 6 Weeks

Immediately post-partum, the mare should be left in peace to bond with her foal. She should have a good supply of hay or fresh grass plus clean water available to her. After an hour or so, she may be given a small nutritious feed. She should then be just discretely observed from a distance (Asbury, 1993). Apart from being involved in the management practices applied to the foal, it is important that, during the first few weeks, she is watched carefully to ensure that she has not suffered any detrimental long-term effects of the birth. In general, however, a lactating mare should be managed in much the same way as other stock, with common sense and an eye for potential problems.

16.5.1. Milk production

At about 72 h after delivery, the mammary gland produces predominantly milk rather than colostrum. Details of milk composition and quality, plus further details of the physiology of lactation, are given in Chapters 9 and 10. During the start of lactation, it is important that the mare looks healthy and fit in herself, as any infections or disease will easily pass on to the foal. The general well-being of the foal should also be used as an indication of milk production. If the mare is not producing enough milk, the foal will

appear tucked up, the mare's teats may be sore from continual unsuccessful suckling and she may begin to object to the foal suckling, risking the development of a perpetual situation making the condition worse. Low milk yields may be due to a physical inability, low nutritional intake, poor body condition or mastitis (McCue, 1993).

Occasionally, immediately post-partum, the mare fails to produce any milk at all. This is due usually to a failure either in the milk ejection reflex or in the production of milk. Failure of the milk ejection reflex is most common, and is thought to be due to high circulating adrenalin levels as a result of stress and anxiety, which inhibits the action of oxytocin on the udder. Injection of oxytocin and/or warm compresses applied to the udder, along with a quiet calm atmosphere, will often rectify the situation. Failure of the udder to produce any milk, i.e. galactopoiesis or lactogenesis failure, is a serious condition, about which little can be done. Many foals born to such mares have to be classified as orphan foals and brought up accordingly. Such mares should not be bred again, as the condition invariably repeats itself (McCue, 1993).

Teat abnormalities can also cause problems. Inverted or conical nipples make it very hard for the foal to suckle, though the condition may not affect milk production itself. Supernumerary teats may also be present, but are rare, and do not seem to affect milk ejection from the normal teats. Fluid collection occasionally occurs within the udder of high-yielding mares. Udder oedema, as it is termed, results in fluid accumulation around the udder and along the abdomen. As a result the udder becomes too painful to allow the foal to suckle and the condition predisposes the mammary gland to infections.

There is a relatively rare condition in which the mare becomes isoimmunized against the foal; that is, she has raised antibodies against the foal. This results in a fatal reaction within the foal if it ingests the mare's colostrum. Alternative colostrum must be fed for at least 36 h post-partum until the gastrointestinal system loses the ability to absorb

large protein molecules. From this time onwards, the foal can suckle normally.

Mastitis, an infection of the mammary gland, is relatively rare in the mare compared with the cow. It is characterized by a hot, swollen and painful udder, with oedematous swellings developing along the abdomen and up between the hind legs. Milk secreted tends to be thick and clotted and should not be fed to foals. If mastitis does occur, it is most often evident after weaning, especially if the mare is still producing large amounts of milk. Mares that have had foals prematurely weaned are therefore at particular risk. Treatment is similar to that in cows, by the administration of antibiotics directly into the mammary gland via an intramammary tube inserted into the streak canal and repeated for several days. Alternatively, intramuscular injection of systemic antibiotics can be used, as many mares violently object to having a mastitic mammary gland touched.

16.5.2. Uterine involution and breeding on the first oestrus post-partum (foal heat)

Many studs perform a routine internal examination within 3 days of parturition to identify problems and check that uterine involution is progressing appropriately. Any problems identified at this stage can be treated in time for either covering at the foal heat (first oestrus post-partum) or, if not, by her first return to service.

The rate of uterine contractility and involution to its pre-pregnancy state has a significant bearing on the conception rates at covering on the foal heat. Within 7 days, the uterus should have returned to two to three times its size in the barren mare and, by day 30–32, both the uterine body and horns should be back to their normal pregravid size (Blanchard and Varner, 1993a). In addition, recovery of and return to the pregravid state of the endometrium is required; this is normally complete by day 14 (Gygax *et al.*, 1979; Blanchard and Varner, 1993a). Clearance of all uterine fluid discharge should also have occurred by day 15

(McKinnon *et al.*, 1988a). In order for conception rates to be maximized, full uterine involution must have occurred. However, the mare invariably returns to oestrus and ovulation between 4 and 10 days post-partum and, as such, she may be covered before full uterine involution has occurred, depressing conception rates (Belling, 1984). A correlation between uterine involution and conception rates is clearly documented, and mares that return to oestrus and ovulation 10 days post-partum or later have significantly better conception rates to that oestrus, compared with those returning nearer to day 5 post-partum (Loy, 1980). The time interval of 10 days prior to ovulation, plus 5 days for the embryo to reach the uterus, allows 15 days for the uterus to recover prior to receiving another embryo. As has been indicated, by day 15, uterine fluid clearance and endometrium recovery should be complete.

Ideally, mares returning to oestrus prior to day 10 post-partum should not be covered but left until their second oestrus post-partum. However, the industry demands that foals are born as early in the year as possible and that a mare should produce one foal per year. The mare's 11-month gestation makes this difficult to achieve. In practice, there are several management techniques that can be used to help achieve these aims. The main aim is to manipulate the timing of the first or second ovulation and oestrus, in order to allow a 15-day recovery period prior to the arrival of the embryo in the uterus. The mare's foal heat can be delayed, using progesterone supplementation for 10 days post-partum followed by withdrawal. This will induce a mare to ovulate within 3–4 days. Thus the minimum 15-day recovery period is achieved. Alternatively, the mare may be allowed to show her normal post-partum oestrus and ovulation. If this occurs prior to day 10, she is not covered but her next ovulation is advanced by the use of prostaglandin $F_{2\alpha}$ 6–10 days after the foal-heat ovulation, again ensuring at least a 15-day recovery period. Both these methods allow longer time for uterine involution and hence result in better conception rates (Blanchard and Varner, 1993a).

16.5.3. Nutrition

One of the specific areas to note in the management of the lactating mare is her nutrition. A lactating mare has higher nutritional requirements than any other equid, even one in heavy work (Doreau *et al.*, 1988). As parturition approaches, her nutritional demand increases to meet the demands of the growing fetus. After parturition, the mare continues to provide all the nutrients for that foal, but now the foal is extra-uterine and is larger. The supply of nutrients via milk is less efficient than nutrient transfer via the placenta. The efficiency of energy transfer in milk is only 60%, i.e. 40% of the energy intake does not appear as energy in the milk (Tables 14.2 and 16.3–16.5). After parturition, the mare's nutrient requirements increase by 70–75% during early lactation and by 50% in later lactation, to make up in part for this

decrease in efficiency (McCue, 1993). At peak lactation, a mare may produce up to 3% of her body weight as milk.

16.5.3.1. Protein and energy

Protein and energy are important components of a lactating mare's ration. If her diet is nutritionally low in protein, she will not have enough to satisfy the demands of her milk production. Milk production will then decline (Martin *et al.*, 1991). Low dietary energy will result in mobilization of the mare's own body reserves in order to try and maintain production. If low dietary energy persists, milk yield will decline and the mare will become emaciated (Pagan and Hintz, 1986). Some weight loss, especially in mares that milk well, is to be expected, but this should be minimized by appropriate feeding. Weight loss in mares that are to be returned

Table 16.3. Daily nutrient requirements of lactating mares of varying weights. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Lactating mares	Weight (kg)	DE (Mcal)	Crude Protein (g)	Lysine (g)	Ca (g)	P (g)	Mg (g)	K (g)	Vitamin A (10 ³ iu)
Foaling–3 months	200	13.7	688	24	27	18	4.8	21.2	12
Foaling–3 months	500	28.3	1427	50	56	36	10.9	46.0	30
Foaling–3 months	700	37.9	1997	70	78	51	15.2	64.4	42
Foaling–3 months	900	45.5	2567	89	101	65	19.6	82.8	54
3 months–weaning	200	12.2	528	18	18	11	3.7	14.8	12
3 months–weaning	500	24.3	1048	37	36	22	8.6	33.0	30
3 months–weaning	700	32.4	1468	51	50	31	12.1	46.2	42
3 months–weaning	900	38.4	1887	66	65	40	15.5	59.4	54

Table 16.4. Nutrient concentrations in total diets for lactating mares (100% dry matter (DM)). Values assume a concentrate feed containing 3.3 Mcal kg⁻¹ and hay containing 2.0 Mcal kg⁻¹ DM. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Lactating mares	Diet proportions			Crude protein (%)	Lysine (%)	Ca (%)	P (%)	Mg (%)	K (%)	Vitamin A (iu kg ⁻¹)
	DE (Mcal)	Conc. (%)	Hay (%)							
Foaling–3 months	2.60	50	50	13.2	0.46	0.52	0.34	0.10	0.42	2750
3 months–weaning	2.45	35	65	11.0	0.37	0.36	0.22	0.09	0.33	3020

Table 16.5. The expected feed consumption by lactating mares (% body weight). (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Mares	Forage	Concentrates	Total
Early lactation	1.0–2.0	1.0–2.0	2.0–3.0
Late lactation	1.0–2.0	0.5–1.5	2.0–2.5

to the stallion can affect conception rates (Sutton *et al.*, 1977; Gill *et al.*, 1983). Hence, the protein and energy content of a lactating mare's ration are very important to her reproductive efficiency. For a 500–700 kg mare, DE intakes of 28–38 Mcal day⁻¹ (2.6 Mcal kg⁻¹ (100% dry matter (DM))) and crude protein intakes of 1400–2000 g day⁻¹ (11–13.2% (100% DM)) are required in order to ensure adequate nutrition for maximum milk production (Tables 14.2 and 16.3–16.5.) (National Research Council, 1989; Frape, 1998).

16.5.3.2. Calcium and phosphorus

In addition to protein and energy, calcium and phosphorus intakes are also very important. For a 500–700 kg mare, average calcium intakes of 56–78 g day⁻¹ (0.52% (100% DM)) are required to ensure that the foal obtains adequate calcium for bone and tendon growth and development. As previously discussed, the ratio of calcium to phosphorus is important; excess phosphorus causes a drain of calcium from the mare's bones in an attempt to redress the balance. Phosphorus intakes of 35–50 g day⁻¹ (0.3% (100% DM)) are required in early lactating mares of 500–700 kg.

In order to satisfy the demands for milk production, the mare will require concentrate feed in addition to good-quality forage. Phosphorus and protein tend to be deficient with regard to lactating mares, even in very good-quality forage diets. The concentrate ration should address this but consist of no more than 50% of the total diet. Not only should protein quantity be high to account for the inefficiency in conversion of nutrient protein to milk protein, but protein quality is also important (Frape, 1998). Lysine is often a limiting essential amino acid in conven-

tional grain and grass forage diets. This can be addressed by the inclusion of lucerne, or soybean meal (Frape, 1998).

As stressed in earlier discussions, it is imperative that all home-grown or bought-in straights and forages are analysed. An appropriate ration for a lactating mare can be ensured using this information and an analysis of commercial concentrates (Tables 14.2 and 16.3–16.5).

16.5.3.3. Vitamins

Extra vitamins and minerals are also required during lactation. Deficiency will cause lacklustre and ill thrift in both the mare and the foal and inefficient use of other nutrients. Vitamins A and D are of particular importance. Vitamin A is available in fresh green forage and vitamin D from exposure to sunlight (Frape, 1998; Duren and Crandell, 2001). Lastly, but by no means least, free access to clean fresh water at all times is essential as 90% of milk is in fact water.

It is not appropriate to be prescriptive on exactly what a lactating mare requires, as requirements will vary with individuals. Mares that produce more milk will probably require more feed and higher levels of concentrate to meet their demand. However, some mares seem to be better doers and more efficient as milk producers. In such mares, obesity may prove to be a problem and, therefore, limiting their concentrate intake should help. The yardstick to go by is the body condition of the mare; she should be fit, not fat, with a body condition score of 3. From 6 weeks of lactation onwards, the foal, as far as nutrition is concerned, becomes increasingly independent. The mare's milk yield then decreases; her nu-

trition management must therefore reflect this change in demand.

16.5.4. Exercise

The importance of exercise in uterine recovery is not to be underestimated. Increasing exercise in the first few days post-partum is linked to accelerated uterine involution and hence conception rates at the foal heat. It is interesting to note that conception rates to the foal heat in feral ponies appear to be relatively normal (Camillo *et al.*, 1997). Confinement of domesticated mares may therefore be a contributory cause of decreasing foal-heat conception rates (Lowis and Hyland, 1991).

16.5.5. Immunization, parasite control, dentition and feet care

No specific immunization of mares that were appropriately vaccinated before parturition, apart from annual boosters, should be required at this stage. Worming should be carried out in the first week, especially against strongyles and ascarids, which are particularly significant in mares and young foals. From then on, a normal regular worming regime should be adhered

to. Teeth and feet care should not be neglected and their care should be continued as normal.

16.6. Foal Management – 6 Weeks of Age Onwards

From 6 weeks onwards, the foal becomes increasingly independent of its mother.

16.6.1. Exercise

Continued turnout is essential to help muscle coordination and development, fitness, gastrointestinal and cardiovascular system function and independence. Ideally, mares and foals should be turned out together in groups to help the foal's development of social awareness and appreciation of hierarchy. Group turnout is an ideal starting-point for gradual weaning systems.

16.6.2. Handling

The foal's handling at this stage should develop from that started in the first 6 weeks of life, remembering patience and reward. Halter breaking should have been started by now (Fig. 16.17). Late halter breaking can lead



Fig. 16.17. By 6 weeks of age, halter breaking should have been started (Penpontbren Welsh Cob Stud).

to confrontation. Leading lessons should develop and the foal should learn to be led without resistance. The process can be aided by a rope around the foal's hindquarters, which can be pulled to encourage it to walk forward. Well before weaning, the foal should be happy to be led without resistance or fuss, both behind its mother and away from her. This can only be achieved by continual and patient training, using short and frequent lessons (Fig. 16.18). The foal should also become further accustomed to travelling, leading on to travelling alone.

16.6.3. Feet care

Foot problems can be identified and possible correction considered within the foal's first year of life. In addition to regular trimming and handling, this can ensure that minor faults and problems can be identified before training begins. Overzealous attack of a foal's feet, in attempts to correct leg problems, should be avoided, as it exacerbates existing problems. Indeed, if left alone, many such deformities often prove to be self-correcting.

Corrective trimming should be done only by a trained and experienced farrier or veterinary surgeon. Deformities, such as an incorrect hoof: pastern angle can be corrected within reason by specific trimming to change the length of the horse's heel. Toes out or in

result in uneven wear of the hoof wall, and corrective and compensatory trimming can alleviate the problem. Excessively long toes or the opposite – club-foot – can be corrected by ensuring that any trimming done is adequate, not overzealous.

16.6.4. Behaviour

From 6 weeks of age the foal continues to develop its independent traits, spending more and more time away from its mother and playing and interacting with other foals. This is invaluable in developing social awareness and learning about hierarchies within a group, in preparation for survival alone with its peers and without its mother for protection.

16.6.5. Nutrition

From 6 weeks of age, a creep feed becomes increasingly important to the foal as a source of nutrients. From this stage onwards, milk quality declines as an encouragement to the foal to seek nutrients elsewhere. The quality of creep feed fed must be carefully assessed to ensure that it provides all the nutrients required for optimum growth and development. However, it should be borne in mind that optimum growth is required, not maximum growth.



Fig. 16.18. The foal should learn to be led without resistance from an early age (Penpontbren Welsh Cob Stud).

As a rough guide, a foal destined to make 150–160 cm may gain up to 2 kg day⁻¹ but by 1 year of age it should not weigh in excess of 80% of its expected mature weight. Excess weight gain causes strain on muscles, tendons, joints, the circulatory system, etc. It is especially important when these structures are still developing that undue stress does not cause permanent deformity.

The protein requirement of the foal for growth is high and it becomes the first limiting factor as far as nutrients supplied by milk are concerned. Creep feeds have been discussed previously (section 16.3.5), but, for the older foal, the feed should contain 20% protein in a highly digestible form; 0.8% should be calcium and 0.6% phosphorus, though a calcium:phosphorus ratio within the range of 1:1 and 3:1 is acceptable. The importance of zinc and copper is increasingly evident.

As far as quantity of feed is concerned, 0.5 kg day⁻¹ at 2 months is adequate, gradually increasing as weaning approaches. As a guide, foals of 3 months of age should be consuming 0.3 kg 100 kg⁻¹ body weight day⁻¹, which is normally about 1 kg day⁻¹ or 0.25–1% of body weight (Frape, 1998).

It is important that the mare does not have access to the foal's creep feed, as this may discourage the foal from feeding. Specially designed creep feeders permit the foal to feed alone without danger that the mare can have access to the feed. The amount of feed per foal should also be controlled and, if several

are run together, it is best if they are fed individually. This may prove difficult but will ensure that each foal is fed according to need and monitored against weight gain. Free access allows greedy foals to gorge themselves at the expense of smaller, less dominant individuals.

If foals are fed outside in a creep feeder, then food should be checked regularly to avoid mould developing. Free access to fresh grass or alfalfa is essential, and access to a mineral supplement is good practice.

16.6.6. Dentition

By 6 weeks, the foal's first and second incisors should have erupted. These are then followed by the wolf-teeth at 5–6 months and then the third deciduous incisors at 6–9 months (Table 16.6). Attention to teeth beyond familiarization with opening the mouth to allow the teeth to be viewed should not be required at this stage.

16.6.7. Immunization and parasite control

Immunization against strangles or rabies may be considered at this time, depending on the prevalence of the diseases. As discussed in section 16.3.7, the best protection against the more common diseases, such as tetanus and influenza, is provided by colostrum.

A regular worming regime should be

Table 16.6. The ages of eruption of equine teeth (H. Tremaine, 2002, personal communication, Wales).

Tooth	Deciduous	Permanent
1st incisor	< 1 week	2.5 years
2nd incisor	4–6 weeks	3.5 years
3rd incisor	6–9 months	4.5 years
Canine	–	4–5 years
Wolf-tooth	5–6 months	
1st cheek-tooth	Birth–2 weeks	2.5 years
2nd cheek-tooth	Birth–2 weeks	3 years
3rd cheek-tooth	Birth–2 weeks	4 years
4th cheek-tooth	–	9–12 years
5th cheek-tooth	–	2 years
6th cheek-tooth	–	3.5–4 years

established from 2 months of age. Wormers against strongyles and ascarids are particularly important, as these specifically affect young horses (Fig. 16.19); Rossdale and Ricketts, 1980).

16.7. Management of the Lactating Mare – 6 Weeks Post-partum Onwards

From 6 weeks post-partum, lactation yield decreases, along with the quality of milk produced. The mare's udder shrinks as milk demand reduces.

16.7.1. Nutrition

As the mare's milk yield decreases, so does her nutrient requirements. Concentrates should therefore be slowly reduced to ensure that she does not become obese. For a 500–700 kg mare, protein intakes can be gradually reduced to 1000–1500 g day⁻¹ (10–11% (100% DM)) and DE to 24–32 Mcal day⁻¹ (2.5 Mcal kg⁻¹ (100% DM)) by 3 months post-partum. Calcium and phosphorus are still important, intakes of 36–50 g

day⁻¹ (0.36% (100% DM)) and 22–31 g day⁻¹ (0.22% (100% DM)), respectively, being required (Tables 16.3–16.5). Particular attention should be paid to the mare's body condition, as mares often put on extra weight in the summer months as the demand for milk declines. It is not appropriate to put a mare in foal on a strict reducing diet later on in the autumn in an attempt to lose excess weight gained during the summer months.

16.7.2. Exercise

Exercise at this stage is essential to build up the mare's fitness again. Indeed exercise, along with a decrease in nutrient intake, helps to dry up milk production. The mare and the foal should be turned out for as long as possible, ideally day and night, providing the weather is good. If the weather is too hot it may be appropriate to bring them in during the day to avoid the flies and turn them out at night.

16.7.3. Immunization, parasite control, teeth and feet care

Worming regime, immunization programmes and attention to teeth and feet should be maintained and not neglected, to ensure that the mare remains in optimum condition, especially if she is in foal again.

16.8. Conclusion

In summary, the management of the lactating mare and foal is particularly critical during the first few hours post-partum. Once this initial critical adaptation of the foal to the extra-uterine environment has successfully been achieved, the likelihood of the foal surviving is high. From this stage onwards, concentration should be given to the management of the mare so that she can be successfully covered as soon as possible, and to the management of the foal so that it grows into a healthy, well-disciplined individual.



Fig. 16.19. Regular worming should be established by 2 months of age and continue as a routine for the rest of the foal's life.

17

Weaning and Management of Youngstock

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17.1. Introduction

Correct weaning and youngstock management is critical in ensuring the foal's long-term good health, physical growth and development, psychological development, social interaction with fellow equids and with humans and long-term productivity. The following account will consider weaning

management and the management of young stock separately.

17.2. Weaning

Weaning is essential to allow the mare's mammary gland to recover in order to ensure an adequate milk supply for the forthcoming,

new foal. Naturally, the foal would be weaned at 9–10 months of age, giving the mare at least a month to recover before the birth of the new foal. By 9 months of age, the foal will normally be consuming a large quantity of solid food, with minimal reliance on milk. The resultant dry period, after weaning and before the new lactation, allows the mare's system to recover and to concentrate on supporting the foal *in utero* and replenishing body reserves.

17.2.1. The timing of weaning

Naturally, during the last 3 months of lactation, the foal, now over 6 months of age, derives most of his nutrients from grass and herbage. It is therefore quite possible, with careful management and the provision of a concentrate supplement feed (creep feed), to wean foals at 6 months, providing they are consuming adequate amounts of feed. Weaning at 6 months is therefore the practice in most studs. However, it should not be considered unless it is certain that the foal is in good physical condition, as well as taking in adequate concentrates. The time of weaning will also depend upon the mare's behaviour, the month of parturition and the dependence of the foal upon the mare (Apter and Householder, 1996). Early weaning, as soon as 4 months of age, may be practised if the mare is suffering. This requires planning and good management, but, providing the foal is well prepared, is introduced to solids soon enough and is in good physical condition, it should not suffer significantly as a result. Early weaning can cause complications, as such foals cannot be run out with other foals, which will either not yet be weaned or will dominate the foal and not allow it adequate access to concentrate feeds. An alternative companion needs to be provided to give psychological security and development; a small, quiet donkey or pony can be ideal.

17.2.2. Weaning stress

Weaning is a very stressful process, both physically and psychologically. The foal will

be separated from its dam, milk will be eliminated from its diet, it will be introduced to strange horses and there will more handling and contact with humans. Careful management, however, can ease these stresses and thus reduce the stress of weaning.

The physical stress of weaning can be reduced by ensuring that the solid-food intake of the foal prior to weaning is adequate to minimize any setback due to the sudden removal of milk from the diet. Sudden changes in diet at all ages can cause digestive upsets and in foals they can additionally lead to growth and developmental retardation (Warren *et al.*, 1998; Fig. 17.1). To ensure a gradual change, some studs advocate milking the mare for a few days after weaning and feeding the milk to the foal along with its concentrate diet. This is also reported to reduce the risk of mastitis but is time-consuming and some mares will object violently to being milked by hand. Alternatively, foals can be fed milk pellets for a while after weaning. Such artificial inclusion of milk into a weanling's diet, however, defeats one of the main objects of weaning – that of removing milk from the diet. In addition, if the foal has naturally significantly reduced its intake of milk prior to



Fig. 17.1. Intake of solid food must be adequate prior to weaning so as to minimize the upset to the digestive system as a result of the change from a liquid, milk-based diet to a solid, concentrate- and forage-based diet.

weaning, the addition of milk after weaning will be a retrograde step and may adversely affect newly established gut microflora and so cause digestive upsets.

A foal being considered for weaning must be in good health. Any animals showing signs of illness, such as a runny nose, coughing, listlessness, a starry coat, diarrhoea, etc., must not be weaned until their condition improves. Young animals can suffer quite dramatically from seemingly small problems, resulting in considerable setbacks to their development, with possible permanent damage. If in doubt, it is advisable to call a veterinary surgeon. In exceptional cases, and only under veterinary supervision, foals suffering from ill health may be weaned, as some medicines are easier to administer and more effective in a foal that is not on a milk diet. Psychological stress can be reduced by introducing the foal to its post-weaning companions and regular handling prior to removal from its mother (see Chapter 16).

17.2.3. Methods of weaning

Plans for weaning foals should be considered well in advance of the actual event. There are four main types of weaning: sudden or abrupt; gradual; interval or paddock;

and weaning in pairs. The method employed is often dictated by the facilities available, the numbers of foals and youngstock and also personal preference. Traditionally, foals were weaned suddenly and individually by removing the mare abruptly and leaving the foal in a stable or loose box out of earshot of its mother (Fig. 17.2). More recently, other methods have been advocated, which are based on a more gradual removal of the mare or the introduction of substitute companions.

17.2.3.1. Sudden or abrupt weaning

Sudden or abrupt weaning involves the abrupt separation of the mare and foal. If this system of weaning is to be employed, then a safe and secure stable is required for the foal. It must be free from any projections likely to cause damage; water buckets should either be fixed to the wall or not left unattended. Hay should be fed in a hay rack off the ground – a hay net is not advised as the foal may strangle itself; hay on the floor is better but can be wasted. The bed should be deep, ideally made of straw, providing good protection as the foal launches around the box. The stable door should be secure, with an upper grill or metal-mesh door, as well as a solid upper door (Fig. 17.3).



Fig. 17.2. Traditionally foals are weaned by sudden removal of the mare, leaving the foal in a secure stable.



Fig. 17.3. The stable used to leave the foal in after removal of the mare should be very secure and have an upper grill or mesh as well as a solid upper door.

At weaning, the mare is abruptly removed from the loose box, leaving the foal behind. The foal should by now be accustomed to handling and can be held in the stable while the mare is removed. She must be kept moving even though she is likely to be very reluctant; the quicker she is removed and with the least fuss, the better. As soon as she is out of the stable, both solid doors top and bottom should be shut and a light left on in the stable. The foal should be relatively safe under these conditions for a short while until the mare has been attended to.

The mare should be taken to a field out of earshot of the foal, with limited grass cover. The field should be secure, with safe boundaries. Some mares are very disturbed for the first few hours and can easily damage themselves by careering around; others appear to consider weaning a relief. The mare should be watched until she has settled down and started to graze. The foal should then be checked. It should be given water, as it will have invariably worked itself into a good lather. Hay should also be made available *ad libitum*, along with a small feed fed as soon as it has calmed down. The foal should remain in the box for the first few days to allow it to get used to life alone. A large stable is therefore advantageous. For these first few days, the upper mesh door should be closed at all

times to prevent the foal attempting to jump out and yet still providing ventilation. Foals are notoriously unaware of danger and will launch themselves at obstacles that an adult horse would not dream of attempting. They are therefore very prone to damage and extra care should be taken to avoid potential hazards: prevention is infinitely better than cure. The foal should be handled and mucked out regularly.

These first few days are very stressful and the foal is susceptible to physical damage and disease, and this is one of the main disadvantages of this system. After a few days, providing the foal is calm, it may be turned out for short periods of the day with a companion in a small secure paddock. The length of turnout can be increased gradually to all day and night if appropriate. In many systems, foals are still brought in at night right through until the following spring, as weaning does not occur until late summer or autumn.

During this time, the mare should not be neglected. Her udder may start to show signs of tenderness and discomfort due to the increase in milk pressure. The milking out of a small amount of milk daily for the first few days is advocated by some in order to reduce the pressure and hence the chance of mastitis. The amount of milk removed daily should only be small and should grad-

ually be reduced over 5 days. The removal of too much milk will only serve to prolong the problem. If milk build-up within the udder leads to excessive pressure, infection and mastitis can result. Post-weaning mares must therefore be watched for such problems, especially in the first few days. Mastitis in mares is relatively rare but, if it occurs, can prove fatal if not appropriately treated. Antibiotic treatment is usually successful, though there is always the danger of the infected half of the udder being lost.

17.2.3.2. Gradual weaning

Gradual weaning is a newer and increasingly popular method, as it attempts to reduce the stresses of sudden weaning. It can be practised in yards with single mares or groups. As with abrupt weaning, if two stables are to be used for the mare and foal, they must be safe and secure and ideally have an interconnecting barred window. More commonly, adjacent paddocks are used and, providing these are well fenced and secure, there should be no problems. It is advised that the fencing should be post and rail, rather than wire, to reduce the chance of injury. If two paddocks are to be used, it is normal to select the more lush pasture for the foal, as its requirements will be greatest and eating will

provide a distraction. Initially, the mare and foal are turned into the separate paddocks or stables for a short period of time, half an hour or so. Over the next couple of weeks this time of separation increases, until they are turned out separately all the time. The close proximity of the foal to the mare allows physical contact and interaction but does not allow suckling. Independence and a reduction in the reliance on milk are developed over a period of time so that the stress of abrupt complete separation is much reduced. An additional advantage is that, as the time of separation is gradually increased, postweaning mastitis is not a problem.

17.2.3.3. Paddock or interval weaning

Paddock weaning requires careful planning and is not possible with single foals or with foals of vastly differing ages. Ideally, the foals should be born in batches within 2 weeks of one another and brought up together in the same paddock after the first couple of weeks (Fig. 17.4). This allows them to become accustomed to each other and a hierarchy is developed while their mothers are still around to dilute any aggression. In such systems some mares may be seen allowing a foal other than their own to suckle, providing her own foal is not also



Fig. 17.4. In a paddock or interval weaning system, groups of mares and foals of similar ages are run together in preparation for the gradual removal of the mares at weaning.

demanding milk. Near the projected time of weaning, all foals should be checked for physical condition and adequate solid food intake. In an ideal system on large studs, there will also be other batches of younger foals born later and following behind, so any foals not ready for weaning in one batch can be transferred to the next one, giving them 2 more weeks or so to become prepared.

Once all the foals are ready for weaning, the most dominant mare or the dam with the most independent foal can be removed on one day, followed by the next dominant the next day, etc., until all have been removed. Occasionally, a gentle dry mare may be introduced as a companion. The mares must be taken well away from the foals and out of earshot. In this system, the foal that has lost its mother soon forgets, due to the solace and security of its fellow foals and the other mares. Mares should again be watched for mastitis.

17.2.3.4. Weaning in pairs

A final alternative, not used widely in practice, but an area of some research, is paired weaning. Based upon the sudden weaning system, foals are weaned abruptly in pairs, rather than as singles, two foals being left in a stable together (Hoffman *et al.*, 1995). However, this system does require the two foals to be 'weaned' from each other at a later stage, which may be as stressful as initial sudden weaning alone would have been.

17.2.4. Variable stresses of weaning systems

As indicated, gradual weaning and paddock weaning are considered the least stressful, as the foals are solaced by familiar surroundings and companions and any change is gradual (De Ribeaux, 1994). This is supported in work by McCall *et al.* (1985, 1987), who, using vocalization as an indicator of stress, suggested the following ranking of weaning methods, based upon the decreasing stress involved: sudden weaning with no creep feed; sudden weaning with creep feed; gradual weaning with no creep feed; and

gradual weaning with creep feed. However, some of these systems do have disadvantages. Some require the foal to be 'weaned' from its other foal companions at a later stage, prolonging the stress. This is of particular concern in paired weaning systems. Indeed, work by Hoffman *et al.* (1995) and Malinowski *et al.* (1990) indicated that paired sudden weaning was in fact more stressful than single sudden weaning, as measured by plasma cortisol levels and foal behaviour. Malinowski *et al.* (1990) also indicated that lymphocyte proliferation to a challenge of concanavalin A was suppressed in foals weaned in pairs, compared with singles. This would indicate a lower disease resistance. This suppression, however, could be improved by human contact. Other systems, such as paddock-weaning systems, are not possible with a single foal or a few foals, as age-group batches are required.

Regardless of what system is used, weaning is stressful and potentially dangerous. Good management, maintenance of familiar surroundings and routines and gradual preparation in diet and handling for the event can reduce this stress considerably. Physical damage to the foal can be minimized by ensuring that it is in safe and secure surroundings and its health promoted by exercise and access to sunlight and fresh air (Holland *et al.*, 1997).

17.2.5. Postweaning care of the mare

As discussed, the mare's udder should be watched for evidence of tenderness due to milk accumulation. In order to minimize this, after weaning she should be turned out into a paddock with limited grass cover and no supplementary feed (Fig. 17.5). This limits her nutritional intake and forces her to exercise in order to obtain what grass she eats. Exercise helps to relieve pressure within the udder and utilizes nutrients that would otherwise be directed towards milk production. Her paddock should be secure and free of hazards, as some mares may be initially quite agitated due to the absence of the foal. The mare usually recovers more quickly



Fig. 17.5. After weaning, mares can be turned out into a paddock with limited grass cover to help dry up their milk production.

from the separation than the foal and, in some cases, may even seem relieved to be free of the extra burden (Holland *et al.*, 1997).

17.3. Management of Youngstock

The management of youngstock in itself could be the title of a book; hence this section will only attempt to give a summary of the principles and main points to consider. The more specialized texts available should be consulted for in-depth details of the systems that can be used (Jones, 1978; Equine Research Inc., 1982; Coldrey and Coldrey, 1990; Knowles, 1993; Britton, 1998; Lorch, 1998).

17.3.1. Nutrition

A significant amount of the foal's development occurs in its first year of life, though it continues until maturity at 4–5 years of age. Nutrition during this period is therefore of the utmost importance. Nutrition must satisfy the body's requirements for growth and development, but not to excess, as this may cause obesity, which in turn exerts extra strain on young limbs, tendons and the circulatory system, leading to conditions such as degenerative orthopaedic disease, osteochondritis dissecans

and epiphysitis (Ralston, 1997). The direct links between nutrition, growth and development and subsequent performance (athletic and conformational) are increasingly evident (Ott and Asquith, 1986).

Prior to weaning, the foal's digestive system progressively adapts to the digestion of solids and roughage. At the time of weaning, the transition from fluids to solids is complete. In general, nutrient deficiencies in youngstock are more critical than in older animals, and so rations for weanlings should be designed with particular care (Traub-Dargatz, 1993a; Cuddeford, 1996; Ralston, 1997). As a general guide, a weanling should be fed 3–4 kg day⁻¹, split into three to four feeds (Ralston, 1997).

17.3.1.1. Energy

The precise energy, protein, vitamin and mineral requirements vary significantly with rate of growth and expected mature weight. The specific requirements for growth being superimposed upon those for maintenance, i.e. if rapid growth is required, then the required nutrient intakes will be greater than for slow or moderate growth (Ott and Asquith, 1986; Tables 17.1–17.3). Energy is the most important component of a youngster's diet. Low energy levels depress growth and, at the extreme, can cause stunting even if the other

Table 17.1. Daily nutrient requirements of youngstock of varying weights. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy of Sciences, Washington, DC.)

Animal	Weight (kg)	Mature weight (kg)	Daily gain (kg)	DE (Mcal)	Crude protein (g)	Lysine (g)	Ca (g)	P (g)	Mg (g)	K (g)	Vitamin A (10 ³ iu)
Maintenance	200	200		7.4	296	10	8	6	3.0	10.0	6
	500	500		16.4	656	23	20	14	7.5	25.0	15
	700	700		21.3	851	30	28	20	10.5	35.0	21
	900	900		24.1	966	34	36	25	13.5	45.0	27
Growing horses											
Weanling 4 months	75	200	0.40	7.3	365	15	16	9	1.6	5.0	3
	175	500	0.85	14.4	720	30	34	19	3.7	11.3	8
	225	700	1.10	19.7	986	41	44	25	4.8	14.6	10
	275	900	1.30	23.1	1154	48	53	29	5.8	17.7	12
Weanling 6 months											
Moderate growth	95	200	0.30	7.6	378	16	13	7	1.8	5.7	4
	215	500	0.65	15.0	750	32	29	16	4.0	12.7	10
	275	700	0.80	20.0	1001	42	37	20	5.1	16.2	12
	335	900	0.95	23.4	1171	49	44	24	6.2	19.6	15
Rapid growth	95	200	0.40	8.7	433	18	17	9	1.9	6.0	4
	215	500	0.85	17.2	860	36	36	20	4.3	13.3	10
	275	700	1.00	22.2	1111	47	43	24	5.4	16.8	12
	335	900	1.15	25.6	1281	54	50	28	6.5	20.2	15
Yearling 12 months											
Moderate growth	140	200	0.20	8.7	392	17	12	7	2.4	7.6	6
	325	500	0.50	18.9	851	36	29	16	5.5	17.8	15
	420	700	0.70	26.1	1176	50	39	22	7.2	23.1	19
	500	900	0.90	31.2	1404	59	49	27	8.6	27.7	22
Rapid growth	140	200	0.30	10.3	462	19	15	8	2.5	7.9	6
	325	500	0.65	21.3	956	40	34	19	5.7	18.2	15
	420	700	0.85	28.5	1281	54	44	24	7.4	23.6	19
	500	900	1.05	33.5	1509	64	54	30	8.8	28.2	22
Yearling 18 months											
Not in training	170	200	0.10	8.3	375	16	10	6	2.7	8.8	8
	400	500	0.35	19.8	893	38	27	15	6.4	21.1	18
	525	700	0.50	27.0	1215	51	37	20	8.5	27.8	24
	665	900	0.70	33.6	1510	64	49	27	10.9	35.4	30
In training	170	200	0.10	11.6	522	22	14	8	3.7	12.2	8
	400	500	0.35	26.5	1195	50	36	20	8.6	28.2	18
	525	700	0.50	36.0	1615	68	49	27	11.3	36.9	24
	665	900	0.70	43.9	1975	83	64	35	14.2	46.2	30
Two-year-old 24 months											
Not in training	185	200	0.05	7.9	337	13	9	5	2.8	9.4	8
	450	500	0.20	18.8	800	32	24	13	7.0	23.1	20
	600	700	0.35	26.3	1117	45	35	19	9.4	31.1	27
	760	900	0.45	31.1	1322	53	45	25	12.0	39.4	34
In training	185	200	0.05	11.4	485	19	13	7	4.1	13.5	8
	450	500	0.20	26.3	1117	45	34	19	9.8	32.2	20
	600	700	0.35	36.0	1529	61	48	27	12.9	42.5	27
	760	900	0.45	42.2	1795	72	61	34	16.2	53.4	34

Table 17.2. Nutrient concentrations in total diets for youngstock (100% dry matter). Values assume a concentrate feed containing 3.3 MCal kg⁻¹ and hay containing 2.00 MCal kg⁻¹ of dry matter. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy of Sciences, Washington, DC.)

Animal	DE (Mcal kg ⁻¹)	Diet proportions		Crude protein (%)	Lysine (%)	Ca (%)	P (%)	Mg (%)	K (%)	Vitamin A (iu kg ⁻¹)
		Conc. (%)	Hay (%)							
Maintenance	2.00	0	100	8.0	0.28	0.24	0.17	0.09	0.30	1830
Growing horses										
Weanling 4 months	2.90	70	30	14.5	0.60	0.68	0.38	0.08	0.30	1580
Weanling 6 months										
Moderate growth	2.90	70	30	14.5	0.61	0.56	0.31	0.08	0.30	1870
Rapid growth	2.90	70	30	14.5	0.61	0.61	0.34	0.08	0.30	1630
Yearling 12 months										
Moderate growth	2.80	60	40	12.6	0.53	0.43	0.24	0.08	0.30	2160
Rapid growth	2.80	60	40	12.6	0.53	0.45	0.25	0.08	0.30	1920
Yearling 18 months										
Not in training	2.50	45	55	11.3	0.48	0.34	0.19	0.08	0.30	2270
In training	2.65	50	50	12.0	0.50	0.36	0.20	0.09	0.30	1800
Two-year-old 24 months										
Not in training	2.45	35	65	10.4	0.42	0.31	0.17	0.09	0.30	2640
In training	2.65	50	50	11.3	0.45	0.34	0.20	0.10	0.32	2040

Table 17.3. Expected feed consumption by youngstock (% body weight). (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by National Academy of Sciences, courtesy of the National Academy of Sciences, Washington, DC.)

Animal	Forage	Concentrate	Total
Maintenance	1.5–2.0	0–0.5	1.5–2.0
Young horses			
Nursing foal 3 months	0	1.0–2.0	1.0–2.0
Weanling foal 6 months	0.5–1.0	1.5–3.0	2.0–3.5
Yearling foal 12 months	1.0–1.5	1.0–2.0	2.0–3.0
Yearling 18 months	1.0–1.5	1.0–1.5	2.0–2.5
Two-year-old 24 months	1.0–1.5	1.0–1.5	1.75–2.5

components of the diet are appropriate. For 4–12-month-old youngsters expected to reach 500 kg mature weight with a moderate rate of growth, energy intakes of 14–15 MCal day⁻¹ (2.90 MCal digestible energy (DE) kg⁻¹ (100% dry matter (DM)) are required. This increases at 12 months of age to 18 MCal day⁻¹ (2.80 MCal DE kg⁻¹ (100% DM)) (Tables 17.1–17.3; National Research Council, 1989). These relatively high energy levels can be obtained by

feeding high-concentrate diets; however, an adequate intake of roughage must also be maintained. A ratio of 3:7 roughage:concentrates is ideal if the roughage has a DE content of 2.0 MCal kg⁻¹ DM. This can be achieved by feeding legumes, such as lucerne. Most other roughages have energy concentrations less than 1.0 MCal kg⁻¹ and so either the amount of concentrations fed or the energy concentration of the hard feed should

be increased to compensate. If the amount of concentrate is increased, it should not represent more than 70% of a 12-month-old youngster's diet (Tables 14.2 and 17.1–17.3; National Research Council, 1989; Ott, 2001).

Within these guidelines, the body condition of the youngster should also be monitored and the level of feed should be altered to account for any over- or underweight. Inappropriate energy levels are a prime cause of obesity in youngstock and are also thought to be contributory to conditions such as epiphysitis, structural deficiencies, contracted tendons, etc. in the growing horse (Ralston, 1997; Warren *et al.*, 1998; Coleman *et al.*, 1999).

17.3.1.2. Protein

Protein is a necessity for bone growth and development, in addition to its involvement in energy utilization. For 4–12-month-old youngsters with expected mature weights of 500 kg and moderate growth rates, protein intakes of 720–750 g day⁻¹ (14.5% (100% DM)) are recommended (Tables 17.1–17.3; National Research Council, 1989). The quality of the protein provided, in the form of amino acid composition, is just as important as the quantity. The adequate supply of essential amino acids is extremely important to youngstock. Lysine, usually the most limiting amino acid in most diets, is especially important in youngsters (Ott, 2001). Hence, even if the total protein content of the diet is appropriate, the horse may still suffer from protein deficiency due to the lack of an essential amino acid. Feeding quality protein straights can alleviate this. Soybean meal is a good source of protein, being highly palatable and high in lysine, and it is a popular choice as a component of a youngster's diet.

As mentioned, protein is involved in the utilization of energy within the diet. Inadequate protein concentrations are associated with signs of energy deficiency, even if there are adequate DE levels within the diet fed. It is therefore usually recommended that protein levels slightly above requirements are fed in order to ensure that energy utilization is optimized (Tables 14.2 and 17.1–17.3).

17.3.1.3. Vitamins and minerals

As might be expected, vitamin and mineral intakes in growing youngsters are of importance. Vitamin and mineral deficiencies within the component feeds of a diet and deficiencies in feedstuffs are due to the environmental conditions under which they are grown, particularly soil deficiencies. For example, grain grown on land deficient in selenium will itself be selenium-deficient. Ideally, all rations fed to youngsters should be analysed for the major minerals, at least for calcium (Ca), phosphorus (P), sodium and chlorine, and, if necessary, an appropriate supplementary feed or a general vitamin and mineral supplement fed as routine to ensure that no deficiencies arise. However, the use of a general supplement may complicate imbalances if adequate levels of specific minerals were originally present within the ration (Frape, 1998; Duren and Crandell, 2001).

17.3.1.4. Calcium and phosphorus

Ca and P are essential for bone, tendon and cartilage growth and are therefore extremely important in a youngster's diet. For a 4-month-old youngster with an expected 500 kg mature weight and a moderate growth rate, respective daily intakes for Ca and P should be 34 and 19 g (0.68 and 0.38% (100% DM)). In practice, levels slightly above this are often fed to allow for a margin of error. Requirements decrease to 29 and 16 g (0.56 and 0.31% (100% DM)), respectively, for youngsters of 6 months of age or older (Tables 17.1–17.3; National Research Council, 1989). However, the ratio of Ca:P is as important as their specific concentrations; excessive Ca reduces P availability and so causes signs of P deficiency. A ratio of 1.5:1 is ideal for 6-month-old youngsters, though some variation either side of this can be tolerated. The older animal is able to tolerate more variation, but, even so, the Ca:P ratio should not exceed 3:1.

17.3.1.5. Sodium chloride

Youngsters' rations should contain 0.9% salt, though the actual demand and intake of salt

will be affected by work and environmental conditions. A salt-lick or general vitamin and mineral supplement containing salt can be made available to youngsters on an *ad libitum* basis to allow them to supplement their salt intake as and when required.

17.3.1.6. Trace minerals

Other minerals required by the growing horse include selenium, iodine, iron, copper, cobalt, manganese, potassium, magnesium, molybdenum, sulphur and fluorine. The requirements are, however, low – hence the term trace minerals. Most feedstuffs will provide adequate levels of these, though certain feedstuffs may be deficient in some trace minerals due to deficiencies within the soil in which they were grown; for example, selenium is deficient in parts of mid-Wales in the UK, in areas around the Great Lakes in the USA and also in parts of New Zealand. If horses are fed exclusively on feeds grown within these areas, they will show signs of deficiencies. Most commercial diets are a combination of feedstuffs from numerous locations and, as such, normally contain adequate trace elements. Problems may be encountered in horses fed purely on home-grown and mixed rations in deficient areas.

A free-access vitamin and mineral lick or routine supplement to the diet can ensure that all the vitamins and minerals required by the growing youngster are provided.

17.3.1.7. Water

Water must not be forgotten as an essential component of any diet; a continual supply of clean fresh water is essential.

17.3.2. Housing

The nutritional requirements of any youngster are partly determined by its environment and housing. The higher a horse's maintenance requirement, the higher its overall nutritional demand. If maintenance requirements are increased by adverse environmental conditions, i.e. wind, rain, cold,

etc., then the overall ration must be increased to compensate. If this is not done, then the growth and development of the horse will suffer. In order to minimize maintenance requirements and feed costs, many youngsters are housed for part, if not all, of the day, especially in adverse weather conditions. This also helps prevent damage and injury to the horse. However, such management is not advantageous to the psychological development of the horse. Isolation can lead to behavioural problems and such horses are also prone to obesity. An alternative that goes some way to solving the problem of isolation is to keep youngsters together in groups in a barn. Youngsters are successfully kept in this way providing time is allowed for them to adjust to each other and to develop a hierarchy, preferably in a large open field, before being confined within the barn. This system does run a higher risk of injury to weanlings from close contact with fellows, and animals kept in this way must be watched closely for signs of bullying and appropriate action taken. Ideally, weather permitting, weanlings should be turned out all the time with supplemented feed. The benefits of social interaction, sunlight and *ad libitum* exercise should not be underestimated.

17.3.3. Exercise

As previously mentioned, exercise and social interaction with other horses are essential for youngsters' physical and psychological development. Ideally, youngsters should be reared at pasture and this is becoming increasingly popular. Such animals can exercise at will, reducing the strain on growing bones and allowing muscles and tendons to develop and grow in strength and size. Social interaction with other horses allows a respect for fellows and reduces boredom and the associated bad habits of confinement (Fig. 17.6). Environmental conditions are often the reason for restricted turnout. Conditions such as heavy rain, snowfall and driving wind or very hot weather may justify youngsters' being brought in. The only



Fig. 17.6. Ideally, youngsters should be reared at pasture, allowing them to develop social interaction and respect for others, as well as providing exercise, to the advantage of bone, muscle and tendon development (Derwen International Welsh Cob Stud).

major disadvantage of pasture-kept youngsters is the difficulty in making sure that all have an adequate intake of concentrates. Most studs feed a standard ration to all pasture-kept youngsters all together and any individuals that appear to be in under- or over-condition are fed separately.

17.3.4. Handling

Before weaning, all foals should be halter-broken and be used to being handled (Fig. 17.7). It is much easier to teach foals basic manners at this age than having to face a stropky and potentially dangerous yearling.



Fig. 17.7. Before foals are weaned, they should be halter-broken and used to being handled, which considerably eases their management after weaning.

A well-handled foal that is used to grooming, feet trimming, clipping, boxing, etc. will be much easier to train in later life. Such an animal will have developed a confidence and respect that can be built upon in later training schedules. It is also much more likely to concentrate on the lesson in hand and therefore be easier to train, as it has confidence in its relationship with its handler.

When handling any youngsters, it must be remembered that they can be very unpredictable. By nature, they are often flighty and there is no guarantee that they will always react to things in the same way. As they develop, their interaction with and therefore their reaction to their environment changes. Every precaution should be taken to ensure that, if the unexpected occurs, no one is hurt and minimum panic ensues. A youngster is very impressionable and panic or nervousness within the human handling it can easily be sensed by and transferred to the youngster. This can become self-perpetuating and result in an owner too frightened to handle the youngster and a horse that is a bag of nerves and increasingly unpredictable in behaviour. A complete outsider is often required to break such a downward spiral.

The youngster should always be treated with constant discipline. It gains security from predictable, consistent handling, even though it will try to push the limits of behaviour. Bad habits and behaviour should be disciplined immediately with the voice. Physical punishment is not normally necessary, except in extremes, and, if not carried out with care, can degenerate into a confrontational situation.

Youngsters should be taught not to bite, nip, push, barge, etc. A foal should not be allowed to play with handlers. It is great to teach a foal to 'shake hands', but, when it starts to kick out in front as a yearling or older horse, this can be very dangerous. In such a scenario, the horse cannot really be blamed, as it will find it very confusing to now be told off for doing something it was once praised for. Charging and pushing handlers, especially when leaving a stable or going through a gate, are potentially very dangerous. The foal should be taught to let

the handler go first and, if it barges, it should be halted with a short sharp tug on the lead rope. If that does not work, then a harder series of tugs should be given. One continual tug is not advised, as this will encourage the horse to fix its neck and head and use its strength against its handler in the future.

Praise is as important as discipline and, whenever a youngster does well or does as it is told, it should be praised by a pat and also by the voice. Throughout its handling, it should get used to the human voice and certain clear commands. Initially, 'Whoa' is very useful to get the horse to stand still and can be used in later lunging lessons. This is one of the most important discipline lessons to learn. As long as a horse will stand still when told, you will always have control over it. There are other commands for other actions, e.g. come to call, walk, trot on, etc., as appropriate. A horse will get used to any commands, providing they are clear and consistently used.

Initially, a youngster will be very exuberant and full of energy and, at this stage, it may be appropriate to overlook minor bad behaviour until the basics have been achieved. It can be very demoralizing for a youngster – and the handler – if it is always being disciplined and can seemingly do nothing right. The less desirable behaviour can be disciplined at a later stage providing it does not get out of hand. The handler must use his or her common sense and assess the situation.

Beyond the basics, further handling and training will depend on the youngster's destination in life and can be geared towards a particular aim, for example, racing, breeding, showing, hacking, etc. It is beyond the scope of this book to discuss the specific training of youngsters for specific disciplines.

Whatever the ultimate destiny for the youngster, exercise is of particular importance. At about a year of age, forced exercise can be introduced in the form of leading out or lunging, and this provides the grounding for further training, especially in riding-horses. Regular exercise also provides conditioning and fittening of horses, ensuring that when training begins in earnest an initial

period of fittening is not required. Training of unfit horses runs the risk of strains and sprains of unconditioned muscles and tendons, leading to a waste of time in resting injuries. A youngster that does not have enough free exercise can be very difficult to train, as it will spend much of the time careering around before work can commence in earnest. Such animals are also very difficult to lunge or put in a horse walker and run a high risk of injury. A combination of free and forced exercise allows agility and physical ability to develop, along with mental stimulation. Development of both mental

and physical faculties is essential as a highly sophisticated state is required in many equine disciplines.

17.4. Conclusion

Considerable thought should be invested in the method of weaning and the handling of youngstock. Management at this young age can have long-term repercussions on the foal's physical and psychological development and so affect its ability to fulfil its potential in later life.

18

Stallion Management

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18.1. Introduction

The management of the stallion should not be neglected in the enthusiasm to obtain optimum mare and foal management. Management of the stallion has already been considered in some depth in Chapters 11, 12 and 13. However, it is also worth considering the introduction of the stallion to his work as a breeding animal and the general training and management principles that should be borne in mind when keeping stallions.

18.2. Early General Training

Early training, well in advance of the stallion's first introduction to a mare, is very important to reduce the chances of injury to both horses and handlers. It will also reduce the risk of him developing potentially dangerous bad habits.

It is very important that a stallion is taught discipline and respect from a young age. Once a good grounding has been established, this can be built upon. It is near

impossible to start disciplining a 3- or 4-year-old stallion without considerable risk of injury, and it will inevitably lead to conflict and not respect. In training a stallion, the handler's attitude and competence are of extreme importance, as rough handling and incorrect and inconsistent training can cause many problems, from poor breeding behaviour and performance to dangerous vices (McDonnell, 2000).

Once basic discipline has been achieved, including the acceptance of the handler, bridle, leading, obeying voice commands to halt, walk on and back up, along with boxing, shoeing, veterinary inspection and general handling, he may be trained further in a specific area or discipline or maintained at this level for breeding. Further training for riding or driving is advantageous, as it provides the stallion with another constructive outlet for his energies other than covering. This advanced level of discipline normally leads to greater respect and subsequently an easier stallion to handle.

18.3. Restraint

There are several means of restraint that can be used to control a stallion. The method used depends on the stallion's age and temperament, the facilities available and the handler's personal preference. The effect of the handler on the behaviour of the stallion cannot be overemphasized. A nervous and insecure handler will transfer these feelings to the stallion, which, in picking them up, is more likely to act uncharacteristically and unexpectedly. It is especially important that the handler dealing with young stallions is calm and confident and has had plenty of experience. The overuse of restraint or punishment to compensate for nervousness is a trap that can, all too easily, be fallen into. If a stallion needs to be reprimanded, it should be immediate, quick and effective. Continuous ineffective, half-hearted attempts, often due to a lack of experience or confidence on the part of the handler, lead to resentment of the handler by the stallion. Stallions are by nature proud and courageous, attributes much to be

admired. They must be treated with care and respect in order to maintain these attributes and channel them into a safe expression and not into conflict (Pickett and Voss, 1975b).

Ideally, the stallion should be restrained for day-to-day management by a good strong leather or webbing head collar or halter and a lead rope. This must be checked regularly as, due to his strength, a stallion may break away easily from inadequate restraint and has the potential to wreak havoc in a yard. He should have been taught acceptance of the halter and leading at a young age so that this is not a problem. However, it is inevitable that at an older age he will become more boisterous and, if he has been inappropriately trained in early life, he may need a more substantial means of restraint. This may consist of a halter plus chain, so arranged over the nose that a pull on the chain applies pressure to the nose and provides extra restraint on the stallion. A more severe method along these lines is to pass the chain through the stallion's mouth or under his chin. This should be done with care, as it is potentially very dangerous for the stallion's mouth and nose if extreme pressure is applied.

Many stallions are restrained, especially for covering, by means of a snaffle or stallion bit and, again, a chain may be attached in one of several positions. It may be passed through the left-hand ring and attached to the right-hand ring (Fig. 18.1); attached to a separate chain that is passed through both rings; or, more severely, passed through the left-hand ring, then through the right-hand ring and passed back to and attached to the left-hand ring. This arrangement of the stallion chain passed under the chin is popular but is thought by some to encourage rearing, and so an alternative is to pass the stallion chain over the nose, looped through the noseband. This is reported to discourage rearing by encouraging the head to come down when pressure is applied (Fig. 18.2). The more severe forms of restraint should only be used as a last resort. Ideally, a stallion should have different tack for varying occasions, so that he knows what is expected of him by the restraint used.



Fig. 18.1. This form of stallion restraint, with the chain passed under the chin, is commonly used but thought by some to encourage rearing.



Fig. 18.2. This alternative form of stallion restraint, with the chain passing over the nose, is thought to discourage rearing, as pressure applied encourages the stallion's head to come down.

The effective use of the handler's voice should also not be underestimated. A clear, confident voice command is just as effective as a physical reprimand to a well-trained stallion. A halter for everyday use and a snaffle bridle for covering, along with the effective use of the voice, are all a well-trained stallion should require (Figs 18.3 and 18.4).

18.4. Introducing the Stallion to Covering

A young stallion should not be expected to cover mares until he is at least 3 years old. Work by Johnson *et al.* (1991) indicates that a 3-year-old stallion is capable of covering mares successfully in his first season, but only a few. A 4-year-old is capable of covering a



Fig. 18.3. All a well-behaved stallion should require for restraint on an everyday basis is a halter (Paith Welsh Cob Stud).

full book of mares (50 per season), but his fertility rates and libido cannot be expected to be consistent. By 5 years of age, he should have reached his full reproductive potential, which, for most stallions, is 50–100 mares per season and up to three mares day⁻¹, with rest periods. The workload that he is capable of at 5 years of age is likely to be that which can be expected of him at least until his 20s, barring unforeseen circumstances. It is advised, therefore, that during his first season a stallion

should be limited to ten to 15 mares spread out over the season and he should not be expected to cover more than one per day. The owner of a young stallion should also be prepared to cancel further nominations in the first season if the stallion is showing signs of losing interest and lacks libido or gets injured. All mares for young stallions should be individually picked, as only mature mares of quiet disposition well into oestrus should be chosen. Such mares are often offered on a no foal, no fee basis, as, even if a semen evaluation has been conducted, the stallion has as yet no proved fertility record.

A stallion's first covering should be with an experienced handler who knows the stallion well. Even if the stallion is eventually to be used in pasture breeding or other non-in-hand breeding situations, it is advisable that his first cover is in hand, or in more controlled conditions, to ensure that evasive action can be taken in the event of emergencies. This first cover is extremely important and its success can seriously affect a stallion's long-term ability and behaviour. It is essential that the mare to be covered is experienced and quiet and in full oestrus. Maiden mares are not advisable as they may be unpredictable and it is enough of a job watching an inexperienced and therefore unpredictable stallion, without having the



Fig. 18.4. A snaffle bridle, along with effective use of the voice, is all a well-mannered stallion should require when covering a mare.

added complication of an unpredictable mare. Ideally, the mare should be slightly smaller than the stallion, making it easier for him to mount (Pickett, 1993d).

The stallion and mare should be prepared for covering as detailed in Chapter 12, though the omission of washing of the genitals is practised by some for the first few covers (Samper, 2000). The stallion should be familiar with the covering area, having been introduced to it beforehand, and extra care should be taken to ensure that the floor is non-slip and that there are no protrusions that may injure him or cause him to fall.

The handler should be experienced in the normal sequence of events when mating mature stallions, as ultimately the novice will need to behave in a similar fashion. An inexperienced stallion cannot be expected to conform immediately. At the first covering, it is best to more or less allow him his head. As discussed in Chapter 12, excessive interference by humans discourages the stallion, and at this stage excessive guidance or discipline should be avoided and the stallion allowed to gain confidence in his ability, before he is taught manners as well. However, potentially dangerous habits, such as kicking or biting, should be corrected immediately, as, if allowed to persist, they could render the stallion unusable.

At his first few coverings, the stallion may need prolonged teasing and may mount the mare several times before ejaculation is achieved. He should not be hurried or forced in any way, as this will only serve to upset him, put him off his stride and result in long-term problems. If he seems unable to ejaculate properly, he should be taken away and returned to his box and tried again with the same mare later on in the day or, better still, with another mare. There is no reason why such hiccups in the first few covers should have any effect on his long-term performance. This first season is all about building up the stallion's confidence in his ability and gradually instilling manners for the sake of safety. It is a gentle balancing act between the two aims, and the rate of progress very much depends on the individual stallion. You must always be prepared to suspend all

attempts at covering if he has a bad experience and to start again at the beginning to restore his confidence. A bad experience may mean he develops an aversion to a particular type of mare, i.e. colour, size, age, etc., or even a permanent reduction in his libido.

The aim of the stallion's first season is to ensure that he associates his new job with pleasure, in a calm and secure atmosphere, so that he will be able to deal with the occasional not so cooperative mare in later life. It is essential that everything is carried out calmly and that any incident is dealt with confidently. Panic and insecurity in the handler will affect the stallion's attitude and performance. Confidence in his handler and surroundings can only serve to enhance his own self-confidence and therefore his ability.

18.5. General Stallion Management

The general management of the stallion is extremely important to ensure that he is fit, able and willing to do his job. It also helps to prevent the acquisition of bad habits and increases safety. The main areas of stallion management that need to be considered are housing, exercise, nutrition, feet care, dental care, vaccination and worming programmes.

18.5.1. Housing

Naturally, a stallion would roam wide areas of land, migrating over new pasture with his mares. He could therefore exercise himself at will and was always provided with fresh clean grazing. Domestication has largely put paid to this, except in some pasture breeding systems. A stallion's management needs to compensate him for this loss and so optimize his welfare and performance.

Ideally, a stallion should be turned out in a large paddock, but this is not always possible. Climate and limited grazing in many areas preclude the use of all-year turnout.

Most stallions are confined to a stable for some period of time at least. This stable must be large, at least 5 m × 5 m for a 150 cm high

horse, and be light and airy. It should have a good strong secure door, with top and bottom sections, plus a top grid that can be shut to provide extra safety but still allow ventilation. Details, such as the stallion's pedigree, can be displayed, adding interest, especially on yards where visitors are catered for (Fig. 18.5). The stable should have a tie ring at the back, to which the stallion should be tied (racked up) as part of his general daily routine, normally when his stable is mucked out and water and hay replenished. It is a good idea to rack stallions up routinely as it makes them easier to handle if visiting mares are around in the yard, and allows them to be easily caught if required. A consistent routine enhances their discipline.

A stallion's stable, as with all horses, should be kept clean and free of flies. Regular cleaning of water troughs and feed mangers is essential.

In order to reduce boredom and the development of vices, such as crib-biting, weaving, stable-walking, etc., the stable door should overlook a busy part of the yard but not be too close to any mares. Chains, plastic bottles and swedes hung from the ceiling, or a football or even a cat have been successfully used to provide the stallion with entertainment and thereby reduce boredom. The box should have easy access to a paddock, which is nor-

mally exclusively for his use. Two acres per stallion is ideal and allows plenty of room for exercise. The cost of fencing can be minimized, as only this paddock need have strong, high, stallion-proof fencing. For larger stallions, fencing should be post and rail, at least 2 m high. An electric fence, run along the top or projecting into the field about 15 cm from the top of the fence, may be added to provide extra security (Fig. 18.6).

The paddock should be cleared of manure at regular intervals to ensure that it remains clean and to reduce the parasite burden. It may also be provided with a field shelter, to give the stallion protection in inclement weather.

Recent research indicates that the housing of stallions has a direct effect upon reproductive performance. Housing stallions in close proximity to other stallions may mimic the natural bachelor-herd scenario (McDonnell and Murray, 1995). Stallions in such bachelor groups have lower testosterone levels and therefore a lower libido than harem stallions. Presumably, it is the social interaction with mares in the absence of other stallions that results in elevated testosterone concentrations. Pasture-bred stallions are reported to have higher libido and exhibit higher fertility rates than stabled, in-hand-bred stallions (McDonnell and Murray, 1995). In general, it

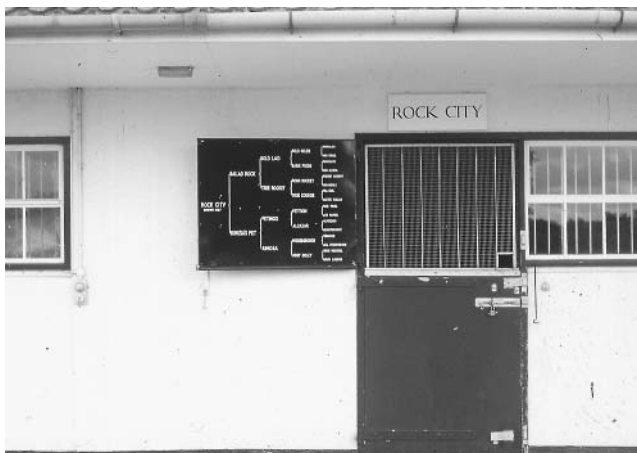


Fig. 18.5. A stallion box at the National Stud, Newmarket, showing the stallion's pedigree displayed on the inside of the upper doors.



Fig. 18.6. Two-metre-high post and rail fencing is ideal for a stallion's paddock. An electric fence running along the top or inside the fencing provides extra security (National Stud, Newmarket, UK).

appears advantageous for stallions to be turned out under natural daylight as much as possible, with plenty of exercise, and, within the constraints of safety, any social interactions should be with mares rather than other stallions, at least during the breeding season (McDonnell, 2000).

18.5.2. Exercise

Exercise, as for all horses, is essential for the physical and psychological well-being of the stallion. It helps to reduce boredom and maintain basic fitness and muscle tone. Fitness is especially important, as stallions undergo short, sharp periods of extreme exercise when covering. Exercise improves the cardiovascular system and reduces the chances of conditions such as azoturia (tying up) and improves general well-being (Dinger and Noiles, 1986a). Exercise also helps aid digestion and promotes a healthy appetite. It can either be free or forced, i.e. turnout or ridden/lunged. Many stallions are unbroken, in which case the only option is free exercise. As such, they should be turned out for as long as possible each day (Fig. 18.7).

Free exercise is fine for those willing to exercise themselves. However, some refuse to move around the paddock or, at the other

extreme, charge around like mad and pace the fence, spending no time grazing and so losing condition. The exercise of these stallions has



Fig. 18.7. Turnout into a field provides ideal exercise for a stallion, weather permitting (Welton Stud).

to be controlled by means of forced exercise, which also improves discipline, especially in nervous and highly strung stallions. Riding and lunging are popular forms of forced exercise. Other forms include swimming (Fig. 18.8), which is particularly beneficial to the cardiovascular system and for lame horses. Treadmills or horse walkers provide an effective means of forced exercise but should be restricted to stallions accustomed to them (Fig. 18.9).

Some stallions, especially native types, can be turned out with mares and foals. This system has the added advantage that mares returning to oestrus after unsuccessful covering can be detected and re-covered by the stallion turned out with them (Fig. 18.10). This system should be confined to use with well-behaved, older stallions, which are well into their working season.

Exercise must be closely monitored, along with nutrition, to ensure that the stallion



Fig. 18.8. Swimming provides good exercise, especially for the cardiovascular system and for stallions with lameness problems.



Fig. 18.9. Treadmills or horse walkers are a good means of forced exercise but should be restricted to stallions that are used to them.



Fig. 18.10. Stallions of a quiet disposition can be turned out with mares, especially towards the end of the season. This relieves boredom in the stallion and allows mares returning to service to be re-covered (Derwen International Welsh Cob Stud).

remains in body condition score 3. He must not be overexerted or he will not have enough energy for the real job in hand.

18.5.3. Nutrition

A properly balanced diet is essential for a stallion's well-being. Each stallion should be fed individually, according to his size, condition, workload, temperament, etc. He should be in a condition score of 3 and his feed should be carefully monitored throughout the year in order to maintain this. One of the major problems encountered with stallions is obesity. Good nutritional and exercise management can prevent this. During the breeding season, the workload, in nutritional terms, of a stallion with a full book of mares is as great as that of a performance horse. As a general rule, a stallion should have a daily intake of 2–3% of body weight; at least 50% of this should be of good-quality roughage (Hintz, 1993b). Young growing stallions should be fed a slightly higher proportion of concentrates, i.e. a ratio of 60 : 40 of concentrates : roughage.

18.5.3.1. Protein and energy

For a 500 kg mature stallion, a daily protein intake of 820 g (9.6% (100% dry matter

(DM))) is recommended, with higher levels for young stallions (Hintz, 1983, 1993b; National Research Council, 1989; Hurtgen, 2000). Daily energy levels of 20.5 Mcal (2.4–2.6 Mcal digestible energy kg^{-1} (100% DM)), similar to those for horses in heavy work, are recommended (Tables 14.2 and 18.1–18.3).

18.5.3.2. Vitamins and minerals

Many breeders feed a vitamin and mineral supplement on a free-access basis to stallions, regardless of feed analysis. This is not always necessary, but can be used as a precaution. The only vitamin that is likely to be short in a well-balanced diet is vitamin A. However, the inclusion of roughage in the diet in the form of leafy green forages, which are high in vitamin A, helps to address this potential shortfall (Hintz, 1993b; Hurtgen, 2000). There is no research that indicates that any single nutrient can improve sperm quality or quantity (Steiner, 2000).

Inappropriate nutrition is one of the major causes of low libido and poor reproductive performance. Correct monitoring of a stallion's condition and adjustment of nutrition and exercise, accordingly, cannot be overemphasized. However, sudden changes to feeding immediately prior to the breeding season can have as detrimental an effect on

Table 18.1. Daily nutrient requirements of stallions of varying weights. DE, digestible energy; Ca, calcium; P, phosphorus; Mg, magnesium; K, potassium. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by the National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Animal	Weight (kg)	DE (Mcal)	Crude protein (g)	Lysine (g)	Ca (g)	P (g)	Mg (g)	K (g)	Vitamin A (10 ³ iu)
Stallion	200	9.3	370	13	11	8	4.3	14.1	9
	500	20.5	820	29	25	18	9.4	31.2	22
	700	26.6	1064	37	32	23	12.2	40.4	32
	900	30.2	1207	42	37	26	13.9	45.9	40

Table 18.2. Nutrient concentrations in total diets for stallion's feed containing 3.3 Mcal kg⁻¹ and hay containing 2.0 Mcal kg⁻¹ of dry matter. (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by the National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Animal	DE (Mcal kg ⁻¹)	Diet proportions		Crude protein (%)	Lysine (%)	Ca (%)	P (%)	Mg (%)	K (%)	Vitamin A (iu kg ⁻¹)
		Conc. (%)	Hay (%)							
Stallion	2.40	30	70	9.6	0.34	0.29	0.21	0.11	0.36	2640

Table 18.3. Expected feed consumption by stallions (% body weight). (Adapted with permission from *Nutrient Requirements of Horses*, 5th revised edn, copyright 1989 by the National Academy of Sciences, courtesy of the National Academy Press, Washington, DC.)

Animal	Forage	Concentrate	Total
Mature stallion	1.0–2.5	1.0–1.5	2.0–3.0
Young stallion	0.75–2.25	1.25–1.75	2.0–3.0

performance as over- or under-nutrition *per se* (Hintz, 1993b).

18.5.4. Feet care

The feet of a stallion should never be neglected. Lameness can severely reduce and restrict the stallion's ability to cover. This is especially evident in hind-leg lameness and is often first evident as an uncharacteristically low libido. A stallion should ideally not be shod during the breeding season, as shoes can inflict more damage than unshod feet. Regular, 6- to 8-week trimming should be

carried out to ensure that the feet remain clean and uncracked. Any problems should be dealt with immediately to avoid long-term complications. Regular turnout is conducive to good-quality hooves, again reinforcing the ideal of keeping stallions in paddocks for at least part of the day (Hurtgen, 2000).

18.5.5. Dental care

Uneven, rough teeth, along with lesions or abscesses, can reduce a stallion's appetite due to pain. They can also cause food to pass

into the stomach without full mastication, reducing the efficiency of digestion. If a stallion starts to lose condition for no obvious reason, the first things to check are his teeth and mouth. In any case, a stallion's mouth should be checked annually to see whether rasping is required.

18.5.6. Vaccination

Vaccination is an essential part of a preventive-medicine routine that should be developed and implemented regularly for a stallion throughout his life. The vaccinations required depend upon the country in which the stallion resides and on the prevalence of various infections. In the UK, stallions should have up-to-date influenza and tetanus inoculations. Vaccination against equine herpes virus 1 is becoming more popular in the UK, and it is also available now for equine viral arteritis (Timoney and McCollum, 1997). In other parts of the world, such as the USA, vaccination for rabies, botulism, eastern and western encephalomyelitis, strangles and Potomac horse fever may be considered. In general, vaccination is advised approximately 60 days before breeding starts, ensuring that any resultant fever does not affect breeding performance (Hurtgen, 2000; Steiner, 2000).

18.5.7. Swabbing

Swabbing of the stallion's genitalia to test for pathogenic bacteria, as detailed in Chapter 19, is increasingly practised. An annual series of swabs is a compulsory requirement for Thoroughbreds and is increasingly demanded by other breed societies. Swabs are normally taken from the prepuce, urethral fossa and sheath at the beginning of each season and if a problem is suspected.

18.5.8. Parasite control

Worming is another essential part of a preventive-medicine routine. As with all worm-

ers in all horses, the key to success is regular use and rotation of the product to ensure that resistance does not develop. High worm counts, like any parasitic infection, cause listlessness and hence low libido and reduced reproductive performance. Worming should be carried out regularly, especially throughout the spring, summer and autumn. Some wormers are reported to cause listlessness and reduced libido, but only for a few days after administration. Bearing this in mind, some studs try to organize their worming regime so that stallions are not wormed at the height of their covering season.

18.6. Stallion Vices

Largely due to the manner of current management practices, many stallions are in danger of developing bad habits or vices, largely due to boredom. Prevention is infinitely better than cure and it is therefore essential that the stallion's management is geared appropriately. A regular routine of work, exercise, feeding, etc. and a stable in an area of the yard where activity can be observed go a long way to achieving this. A frustrated and bored stallion releases his energies and tensions in the only way possible to him, by developing vices. These vices can be harmful to the stallion himself and dangerous to the handler and may also affect his reproductive performance. If prevention has failed and vices have developed, there are certain practices that can help control them and/or their effects.

18.6.1. Crib-biting and wind-sucking

Crib-biting and wind-sucking are primarily caused by boredom and are related, often developing the one from the other. During crib-biting, the horse bites part of the stable structure or other convenient object (Fig. 18.11). It is thought to develop from the horse's natural urge to eat or graze regularly. The condition is exacerbated when feed is delivered in small concentrate meals with limited roughage. Increasing roughage and allowing *ad libitum* availability will certainly



Fig. 18.11. Tell-tale signs of crib-biting in a horse: protruding surfaces, commonly stable doors, showing signs of teeth marks.

reduce the chance of this vice developing and will help alleviate sufferers. The habit can be discouraged by removing all objects that can be grasped by the teeth or by painting structures with Cribox or an equivalent foul-tasting substance as a deterrent. Wind-sucking can develop from crib-biting and, in this more serious condition, the horse, while grasping the projecting structure, arches his neck and gulps in air. If the habit is allowed to continue unchecked, it can lead to colic and reduced appetite, as well as excessive wear and tear on the upper incisor teeth. A muzzle can be used to prevent both vices. A cribbing strap, placed around the horse's throat, preventing the stallion from tensing the neck muscles used in wind-sucking, may also help (Fig. 18.12).

A more extreme method of curing wind-sucking is severing of the neck muscles attaching the hyoid bone to the base of the tongue. Alternatively, the nerves serving these muscles can be severed. Such a procedure will, in the majority of cases, effect a cure and, in the remainder, considerable improvement is obtained, but these are drastic solutions to a problem that is largely avoidable with appropriate management (McGreevy *et al.*, 1995).



Fig. 18.12. A cribbing strap may be used to prevent the stallion from tensing the neck muscles required for wind-sucking (photograph Sebastian McBride).

18.6.2. Weaving

Weaving involves the lateral swaying of the horse's head and neck rhythmically from side to side, often over the stable door. This can cause damage to the forelegs as the horse's weight is repeatedly shifted from side to side. The condition is thought to develop from the horse's natural urge to move continuously, associated with grazing over large tracts of land. A chronic weaver may weave himself to the point of exhaustion. The condition can be alleviated by anti-weave bars over the lower stable door (Fig. 18.13).

Unfortunately, chronic weavers will continue to weave within their boxes. Furthermore, stable-walking may develop, in which the stallion continually paces around his box, seemingly chasing his tail. There is little that can be done to cure this behaviour except to turn the horse out into a paddock to relieve the boredom, but such horses often then fence walk and usually revert to stable-walking as soon as they are stabled again (Mills and Nankervis, 1999).

18.6.3. Self-mutilation

This vice, unlike the others, is not normally an expression of boredom, but nevertheless can be extremely distressing to the stallion and his owner. The stallion bites his own legs, shoulders and chest, causing himself considerable damage. It is normally particularly evident after mating. If evident only then, thorough washing of the stallion after dismount reduces expression of the vice, which in this case is thought to be due to the smell of the mare. If, however, the stallion is a habitual self-mutilator, there is very little that can be done to cure him, though the use of a muzzle or cradle can prevent him inflicting damage.

18.6.4. Masturbation

Masturbation by a stallion is considered by some to be a further vice, thought to originate from boredom, especially if sexually frustrated. However, it is evident that feral and wild ponies also demonstrate such behaviour. It has been suggested, therefore,



Fig. 18.13. During the times that the top door of the stable is open, anti-weave bars may be used to discourage the stallion from weaving over the bottom door.

that it is a natural behaviour, rather than a problem, and any problem lies with human perception and potential embarrassment. The old-fashioned use of penile rings, etc. is now frowned upon and considered unnecessary. Masturbation is expressed by the stallion rubbing the extended penis along the under side of his abdomen. In extreme cases, masturbation may result in ejaculation and concern over the loss of valuable sperm. Providing the stallion's workload is not too high, such behaviour should not affect his fertility rate; indeed, increasing his workload may go some way to reducing it (Pickett, 1993d; McDonnell, 2000).

18.6.5. Aggressive behaviour

Some stallions develop extremely aggressive behaviour and become a danger to both handlers and mares (McDonnell, 2000). Occasionally, this behaviour is associated with certain conditions or restraint or is directed towards certain people and can be averted by avoiding such situations. However, more often than not, it is expressed generally and is due to mismanagement during formative years. If the behaviour is beyond control, the stallion can be gelded. In 95% of cases, gelding significantly reduces aggressive tendencies. If the stallion must remain entire, then certain measures can and should be used to protect handlers and mares. He should be muzzled, to prevent savaging of mares during covering, and controlled by a pole attached to his bit, giving his handler more control. The only option with some stallions may be artificial insemination. In deciding whether to continue to use an aggressive stallion, it must be certain that his behaviour is management-induced and not inherited, as it is very important that such behaviour is not perpetuated in subsequent generations.

Rearing and striking out with the front feet constitute a relatively common vice, though potentially very dangerous to handlers. This should be corrected, especially in

young stallions, where the vice can be cured. To avoid being kicked, the handler should always stand to one side of the stallion, and never in front. A long lead rein should be used so that contact can still be maintained from a distance if the stallion rears. As soon as the stallion starts to rear, his lead rein should be jerked sharply, along with a verbal reprimand. Backing the stallion at the first signs of rearing can also help to avert the situation. The use of a chain under the chin is one of the more popular forms of stallion restraint, but it has been reported to be associated with a higher incidence of rearing and hence may be best avoided.

Finally, biting is another relatively common vice in stallions. This should be corrected at a young age by a short, sharp jerk on the lead rein or a sharp tap on the muzzle and verbal reprimand as punishment. If allowed to continue, a stallion can become almost impossible to handle. Some stallions only bite in certain situations, such as when they are eating, after mating, when they are being groomed or when fed by hand. Such situations should therefore be reduced to a minimum and any handlers warned of the problem.

18.7. Conclusion

Stallion management from a very early age has important implications for reproductive ability and behaviour. Many problems encountered in stallions, which either do not perform to their full potential or exhibit anti-social behaviour, stem from mismanagement at an early age. One of the major problems encountered with stallions is boredom, due to confinement and isolation. This can directly affect libido, performance and other behavioural characteristics. Unfortunately, this often becomes a self-perpetuating downward spiral, which is very difficult to break. Many of the problems encountered in the management of stallions can be averted by consistent discipline and alleviating boredom by providing turnout, social interaction and activity.

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Infertility

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19.1. Introduction

Infertility is a vast subject and this chapter can only be an introduction, providing a basis from which further information can be sought. Infertility may have its root cause in either the stallion or the mare, and each of these aspects will be discussed in turn.

Expected fertility rates vary enormously and are affected, among other things, by breed (Table 19.1). As an average within the UK, 40–80% of mares covered deliver a live foal (Osborne, 1975; Sullivan *et al.*, 1975; Baker *et al.*, 1993). Failure to produce a live offspring in any single year can be due to extrinsic or intrinsic factors, and this is the case with both the mare and the stallion. Extrinsic is the term given to external factors affecting reproductive performance, whereas intrinsic is the term given to internal, usually physiological, factors. When considering the stallion and the mare, these two areas will be dealt with in turn.

19.2. Stallion Infertility

On average, a stallion might be expected to cover one to two mares per day during the breeding season, with a rest day every 7–10 days. This gives reasonable results, with fertilization rates of about 60% (Pickett and Voss, 1975b). These figures are only an average and are affected by the type and condition of mares presented to a stallion and also by the characteristics of the individual stallion. Some stallions are capable of much heavier workloads and some struggle with less (Pickett and Shiner, 1994). It is one of the

major responsibilities of the stallion manager to be aware of the limitations of his individual stallion and to work within these constraints (Kenney, 1990).

Research into the causes of infertility in the stallion is as yet limited, due to previous concentration on the mare. However, in any breeding programme, 50% of the outcome is determined by the stallion and, as such, this deserves fair discussion. The difficulty in obtaining standard figures for fertility and the reluctance of the majority of stallion owners to select for reproductive performance and to assess for breeding soundness have led to relatively low average fertility rates, which have shown little, if any, improvement in recent years. Before discussing the subject further, the following glossary should be noted to prevent confusion in terms.

Sterility permanent inability to reproduce
Infertility temporary inability to reproduce
Subfertility inability, either temporary or permanent, to reproduce at full potential
Impotence temporary or permanent inability to ejaculate semen; sperm capable of fertilizing an ovum may, however, be produced

19.2.1. Extrinsic factors affecting reproductive efficiency in the stallion

Extrinsic factors affecting the reproductive efficiency of a stallion include lack of use, the presentation of subfertile or infertile mares, poor mare management, poor stallion management and the imposition of an artificial breeding season. Each of these factors will be discussed in turn in the context of reproductive efficiency/infertility. It will be noted that many aspects have already been discussed in previous chapters, especially those concerning management, and so such details will not be repeated here.

19.2.1.1. Lack of use

Reproductive efficiency in any animal reflects its use. A stallion may not be used in a particular year by design, due to financial or manage-

Table 19.1. Average fertility rates for various types of horses and ponies (from Day, 1939).

Breed	Average fertility rates (%)
Heavy horses	59
Light horses	52
Thoroughbred	68
Wild ponies	95
Feral ponies	23–80

ment considerations. Disease may also preclude a stallion from use for part or all of a season, due to the risk of direct disease transfer to mares in the case of venereal or contagious diseases, or may limit his ability to perform, in the case of non-contagious disease. Alternatively, the stallion may have suffered from disease or infection during the previous year and is not to be used in the following season in order to allow full recovery or because the long-term effects of disease on his reproductive performance make it inappropriate to use him until he has fully recovered. Diseases of the stallion's reproductive tract will be discussed later, under intrinsic factors.

Finally, semen evaluation is part of good practice and should be carried out regularly at the beginning of each season. Poor semen quality may lead to the stallion being taken out of use until the cause has been isolated and the problem solved.

19.2.1.2. Subfertile or infertile mare

Both mare and stallion are equally responsible for the production of an offspring. A stallion is only as good as the mare he is to cover, and vice versa. It is essential that any mare presented to a stallion is capable of reproducing and does not suffer from any of the factors affecting reproduction that will be detailed in the later sections on mare infertility. If the mare herself is subfertile or infertile, lack of success cannot be blamed on the stallion.

19.2.1.3. Poor mare management

Mare management is discussed in detail in Chapters 12–16. Inappropriate management will adversely affect the mare's ability to conceive and therefore the apparent fertility rates of the stallion that covered her. The most important management area as far as stallion reproductive efficiency is concerned is during the time of covering. Service at an inappropriate time due to a failure to detect oestrus accurately will obviously be reflected in poor fertility rates. Failure to detect oestrus is usually due to either prolonged dioestrus preventing oestrus from being displayed, or infrequent or inaccurate teasing, along with

lack of records and mare observation. In such cases, veterinary examination by means of rectal palpation and/or scanning have been shown to significantly improve oestrus detection and therefore fertility rates.

19.2.1.4. Poor stallion management

Stallion management is discussed in detail in Chapters 12, 13 and 18. All aspects of a stallion's management will affect his ability to cover mares successfully. Stallion management as far as it directly affects reproductive efficiency can be subdivided into the following.

19.2.1.4.1. EXCESSIVE WORKLOAD. As discussed previously (Chapter 18), the amount of work or number of mares a stallion may be expected to successfully cover during a season is highly variable (Pickett *et al.*, 1975c; Pickett and Shiner, 1994). It is one of the responsibilities of the stallion manager to know the capabilities of his stallions. The workload of a stallion depends upon the ability of his testis to produce sperm. Among other things, this is a function of testis size, which can be assessed by callipers or ultrasonically (Fig. 19.1; Love *et al.*, 1991).

Stallions with large testes have larger daily sperm outputs and can cope with a heavier workload than stallions with smaller testes. Testis size is also a function of age, which has an important bearing on fertility, especially at the extremes of youth and old age (Johnson and Neaves, 1981; Pickett *et al.*, 1989; Pickett and Shiner, 1994). It is a good idea, especially with new stallions, to carry out a full semen analysis to give a guide to daily sperm production (see Chapters 11 and 20).

Sperm concentrations are usually in the range of $100\text{--}300 \times 10^6$ sperm ml^{-1} and, for successful fertilization, $300\text{--}500 \times 10^6$ sperm are required (Pickett and Voss, 1972). On average, 50–60% of sperm produced can be classified as normal, progressively motile sperm capable of fertilizing an ovum (Kenney, 1975a; Pickett and Voss, 1975a). The average daily sperm production for a stallion is $600\text{--}2000 \times 10^6$, depending upon season, environment, age, etc. (Pickett and Voss,

Fig. 19.1. The relationship between predicted daily sperm output and testicular volume, for 26 stallions (from Love *et al.*, 1991).

1972). From these figures, it is apparent that, in theory, the average number of successful services a stallion could be expected to perform per day is one to three. There are, however, other considerations to take into account when looking at workloads.

It is interesting to note that total sperm production per week is the same, regardless of whether a stallion is used daily or on alternate days. However, use on a daily basis results in a lower concentration of sperm ml^{-1} (Pickett *et al.*, 1975a). This may be of no consequence in stallions with high daily sperm production rates, as concentrations will still be acceptable, but daily use of stallions with lower daily sperm-production figures may have a detrimental effect on fertility rates.

Excessive workloads may also result in a lack of libido. As a result, the stallion will be slow to breed or may even fail to ejaculate. In such cases, it is best to take the stallion out of work for a short period of time, and reintroduce him a week or so later. If libido is still low, it may well be indicative of further problems.

In an ideal world, a stallion should be used once a day and given regular periods of rest of 1–2 days every 10 days or so.

However, there are many pressures, not least financial, that entice stallion managers to increase workloads. Due to individual variation, some stallions can cope with more than one mare per day or periods of excessive use in a busy season, provided that they are given adequate periods of rest.

19.2.1.4.2. TRAINING MANAGEMENT. As discussed in Chapter 18, early training is of utmost importance for the long-term ability of a stallion to perform to his full potential. A stallion brought up in a relaxed, unstressed environment, with consistent and fair discipline and respect, is much more likely to perform to his full potential in later life.

One of the major problems encountered as a carry-over effect from early life, which is often unappreciated, is stallion isolation. Many managers isolate stallions to ensure the safety of personnel, other stock on the yard and the stallions themselves. This is a self-perpetuating problem, as such treatment often results in boredom, vices and excessive excitability and unpredictability, which in turn result in further isolation in the interests of safety. A happy medium between safety and stallion participation in the general yard activities has to be achieved.

19.2.1.4.3. BREEDING DISCOMFORT. Full physical examination of the stallion is essential before purchase to ensure that no abnormalities are present. Details of this are given in Chapter 11. It is also advisable that stallions undergo regular examinations at the beginning of each season to ensure that no problems have arisen since the last season that may cause pain at breeding. Pain associated with the act of covering can cause a permanent reduction in libido. Poor feet care or conditions such as laminitis cause pain on mounting, especially if the problem is in the hind feet. Muscular or skeletal problems, including arthritis, may also limit the stallion's ability to mount, due to pain. Irritation and soreness of the penis or sheath area may also cause pain at covering, especially if smegma has accumulated or soap or antiseptic wash has not been rinsed thoroughly. Breeding accidents involving inadequate erection at intromission, kicking by a mare and rough handling will discourage a stallion from future covering, as being associated with pain. Finally, when using an artificial vagina (AV), care should be taken that the internal temperature is not too hot, as this will cause pain and will reduce his future willingness, not only to use an AV, but also in natural service (see Chapter 20).

19.2.1.4.4. NUTRITION. As discussed in Chapter 18, appropriate nutrition throughout the year is essential in order to ensure that the stallion is in optimum physical condition for the season. Obese or excessively thin stallions suffer from low libido, and nutrition, along with exercise, is a major determinant of body condition. A body condition score of 3 on a scale of 1–5 is to be aimed for.

As far as specific deficiencies are concerned, only limited research has been carried out. It is known that general, severe, nutritional deficiency is associated with a delay in puberty, testicular atrophy and a reduction in sperm production. Deficiencies in energy and, to a lesser extent, protein have also been associated with low reproductive efficiency (Jainudeen and Hafez, 1993). Severe deficiencies in vitamins A and E and

selenium are specifically associated with a reduction in spermatogenesis in other farm animals and have been suggested in stallions (Ralston *et al.*, 1986). In addition, low dietary intake of copper, iron and/or cobalt results in a reduction in appetite, with accompanying weight loss and anaemia and, via these, a decline in semen quality (Jainudeen and Hafez, 1993).

Obesity will result in a loss of libido and may also cause a reduction in spermatogenesis. Obesity is associated with excess fat deposition within the scrotum, increasing scrotal insulation and hence causing an increase in testicular temperature, with an associated decline in spermatogenic efficiency.

19.2.1.4.5. CHEMICALS AND DRUGS. As discussed in Chapter 12, it is essential that, prior to use, a stallion that has been on a drug regime must be given time to allow that drug to be eliminated from his system.

Anabolic steroids are sometimes used in an attempt to improve male characteristics – for example, weight gain, muscle growth and performance in young horses. They have also been used to improve stallion libido. In humans and other animals, such use of anabolic steroids is known to be associated with infertility and a similar association has been indicated in stallions. Anabolic steroids have been reported to result in a decrease of up to 40% in testicular size and weight (Blanchard *et al.*, 1983; Koskinen *et al.*, 1997). Spermatogenesis is also reduced, with fewer sperm per gram of testicular tissue being produced and lower sperm-motility rates (Squires *et al.*, 1982; Blanchard *et al.*, 1983). Anabolic steroids are therefore not recommended for stallions in breeding work. These drugs not only have an immediate effect, but may also have a long-term effect, at least until they are eliminated from the stallion's system.

Testosterone therapy has been used to improve libido in stallions and is relatively effective. However, it does have serious potential side-effects as far as fertility is concerned. Chapter 4 outlines the fine control and delicate hormonal balance controlling male reproductive functions. If one of the components of the system is altered, it affects

the delicate balance of the whole system and, in the case of testosterone, reduces spermatogenesis (Squires *et al.*, 1997). Testosterone therapy is therefore associated with low fertility due to reduced sperm counts and hence is not advised for use in stallions in work, unless under veterinary supervision.

In addition, the use of any other drug or treatment that causes a drop in appetite, diarrhoea or lack of condition is ill-advised during the breeding season and should only be used under veterinary supervision. Some wormers have also been reported to be associated with a temporary decline in fertility, and many breeders arrange their parasite-control regimes to ensure that stallions are not treated during the breeding season.

19.2.1.5. Imposed breeding season

Reproductive activity in the stallion, as in the mare, is naturally limited by a breeding season, though with enough encouragement most will cover mares out of season (Pickett and Shiner, 1994). As discussed in Chapter 4, season affects the number of sperm per ejaculate, total sperm number, number of mounts per successful ejaculation and reaction times. As a result, fertilization potential out of season is significantly reduced (Pickett and Voss, 1972). The natural breeding season, with its optimum fertilization rates and libido, is nature's way of ensuring that foals are born during the spring and early summer to maximize their chances of survival.

Unfortunately, this natural breeding season does not coincide with the arbitrary breeding season humans have determined in an attempt to achieve foaling as near as possible to 1 January, the official registered birth date of all foals in several breed societies, the Thoroughbred being the most well known. The arbitrary breeding season in the northern hemisphere starts on 15 February, as opposed to the natural breeding season, which starts in April/May. In the southern hemisphere, the imposed season starts on 15 August, as opposed to the natural season in October/November. Stallions are therefore expected to cover mares at a time of the year

when their libido and fertilization rates are naturally low and when they are unable to perform to their full potential. The adverse effect of season on reproductive efficiency is increasingly evident in older stallions (Johnson and Thompson, 1983).

Occasional use at either end of the non-breeding season can be quite successful, but a full workload can in no way be expected. Exact performance depends on the individual animal, but improved fertilization rates and libido can be obtained by the use of artificial lighting in the stallion's stable, from November onwards, to give 16 h light and 8 h dark, so mimicking the early onset of spring and advancing the breeding season (Clay and Clay, 1992).

19.2.2. Intrinsic factors affecting reproductive performance in the stallion

Intrinsic factors affecting reproductive performance in the stallion include age, along with chromosomal, hormonal, physical and semen abnormalities. These will be discussed in turn in the context of reproductive performance.

19.2.2.1. Age

Age is an important aspect in considering the potential fertility of a stallion. Young and old stallions may have problems with taking on a full workload with consistent success.

A young stallion is still learning the job and can easily be adversely affected by his handlers and/or management. He may therefore be slow to breed, mounting several times per successful ejaculation or even failing to ejaculate, ejaculating prematurely or exhibiting enlargement of the glans penis before intromission. Careful treatment and handling during this period are essential to ensure that any such behavioural problems are not perpetuated (Naden *et al.*, 1990). As far as physical capabilities are concerned, puberty (17–22 months) heralds the beginning of sexual activity (Clay and Clay, 1992). Three-year-old stallions may therefore be used for covering and are perfectly capable

of fertilizing a mare, but they have a limited sperm-producing capacity. By 4 years of age, they are capable of producing adequate numbers of sperm to cover as many mares as an adult stallion, but full consistent fertilizing capacity is not attained until 5 years of age, on average (Berndtson and Jones, 1989; Johnson *et al.*, 1991).

At the other end of the spectrum, old age may be a problem. An age-related decrease in semen quality after 20 years of age has been reported by some (Johnson and Thompson, 1983; Amann, 1993a, b), but this is not supported by other work (Johnson *et al.*, 1991). However, a decline in fertility is associated with general age-related problems, such as arthritis, many of which cause pain on mounting, a major cause of low libido and therefore low fertilization rates. If such problems are encountered, they may be alleviated, to a certain extent, by the use of breeding platforms or artificial insemination (AI) as well as allowing him extra time. The effect of age is very variable between different stallions, and older stallions should not automatically be precluded from use, as such animals have had many years in which to prove their worth as far as their own performance and that of their progeny is concerned. Older stallions often tend to be more gentlemanly to handle, know their job well and are good to use on maiden, shy or nervous mares, giving them confidence. When using an older stallion, it is particularly important that his semen should be evaluated regularly and monitored closely to allow a reduction in his workload if a decline in semen quality is detected.

19.2.2.2. Chromosomal abnormalities

Chromosomal abnormalities or genetic inadequacies may be the cause of infertility in stallions that otherwise appear fit. These may be associated with semen abnormalities or more obvious abnormalities of the genitalia. Hermaphrodites, genetic chimeras or mosaics (63XO:64XY), male syndrome (64XX) and Klinefelter's syndrome (65XXX) have been reported in stallions but are relatively rare

(Halnan and Watson, 1982; Bowling *et al.*, 1987; Bowling, 1996; Makinen *et al.*, 2000). Other genetic abnormalities are associated with cryptorchidism (rig) (section 19.2.2.4.1) and testicular hypoplasia (section 19.2.2.4.3), both directly affecting reproductive efficiency (Varner and Schumacher, 1991).

Some genetic deformities may not directly affect reproduction but may preclude the stallion from use, such as umbilical and inguinal hernias. These may well correct themselves naturally, but there is a possibility that the trait will be perpetuated in succeeding generations. Other genetic factors causing abnormalities of the reproductive system will be discussed under the specific areas of the tract detailed below.

19.2.2.3. Hormonal abnormalities

As mentioned previously in this chapter and in Chapter 4, the endocrine control of reproduction is governed by a finely balanced system. Circulating concentrations of testosterone have a direct effect on reproductive performance, on both libido and sperm production; low testosterone levels are often blamed for poor fertility rates (Nett, 1993c). Testosterone, human chorionic gonadotrophin and gonadotrophin-releasing hormone therapy have been used, with mixed success, to address this problem. The lack of success may well be due to the fact that depressed pituitary function is the cause of infertility in only 1% of cases (Boyle *et al.*, 1991; Roger and Hughes, 1991). Abnormal hormone levels may be associated with hypothyroidism, resulting in delayed puberty, smaller testes, decreased spermatozoa production and decreased libido. Feminization of the genitalia may also be observed. It has been postulated that changes in thyroid function may be the cause of stallion summer infertility, associated with elevated environmental temperatures (Brachen and Wagner, 1983).

19.2.2.4. Physical abnormalities

Numerous abnormalities of the stallion's genitalia have been reported. As with most

anatomical abnormalities, they are either caused by disease or inherited. It is reported that one in five males has an anatomical abnormality, the significance of which varies from life-threatening to a minor flaw, which may be of little consequence as far as reproductive performance is concerned but may still reduce his market value. Only those most commonly encountered will be considered in the following sections.

19.2.2.4.1. CRYPTORCHIDISM. A cryptorchid stallion or a rig is an animal in which either one or both of the testes have failed to descend into the scrotum. The passage of the testes from a position next to the kidneys should occur, as a gradual process, *in utero* or during the first few months of life (Fig. 2.9). A cryptorchid stallion may be further classified as abdominal or inguinal, as illustrated in Figs 19.2 and 19.3 (Cox, 1993a,b).

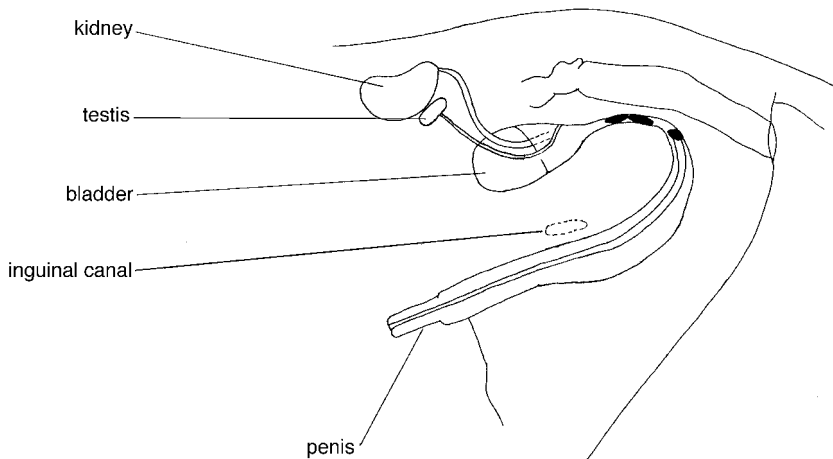


Fig. 19.2. An abdominal cryptorchid stallion is characterized by the testis lying up within the body cavity. In a unilateral abdominal cryptorchid, only one testis has failed to descend; in a bilateral, both remain in the body cavity.

Fig. 19.3. An inguinal cryptorchid stallion is characterized by the testis having only partly descended and remaining associated with the inguinal ring. Again, failure of testis descent may be seen in both (bilateral) or only one (unilateral) testis.

The failure of testes to descend may be temporary (most will descend within 3 years of birth) or permanent. Evidence suggests that more than 75% of cases involve retention of the right testes (Bishop *et al.*, 1964, 1966). Circumstantial evidence and work reported by Cox (1993a) would suggest that the incidence of cryptorchidism is higher in ponies. The retention of one or both testes results in a significant decline in testes weight, even if it does subsequently descend. The size may be reduced by up to 20-fold in the abdominally retained testis; the reduction in size of the inguinally retained testis is not as great but a difference of up to sevenfold has been reported (Bishop *et al.*, 1964). A unilateral cryptorchid is perfectly capable of successfully covering mares, though his total sperm output per ejaculate will be reduced and he will therefore be unable to bear a full workload. In practice, however, cryptorchids are not allowed to be registered for use as a stallion by most breed societies as the condition is heritable.

19.2.2.4.2. HERNIAS. Stallion hernias may be classified in a number of ways (Figs 19.4 and 19.5). All have the potential to affect spermatozoa production, due to an elevation in testicular temperature from the close proximity of the herniated part of the gastrointestinal tract (Cox, 1988).

Testicular hernias may be acquired, usually due to accident or strain (Varner and Schumacher, 1991), or congenital, due to inherited abnormality (Schneider *et al.*, 1982; Cox, 1993a). Testicular hernias may be further classified as inguinal or scrotal, depending on the extent of herniation. Inguinal hernias result from intestinal tissue passing solely through the inguinal ring (Stashak, 1993; Fig. 19.4). Scrotal hernias result from further herniation, where the intestine extends into the scrotum (Varner and Schumacher, 1991; Fig. 19.5). The most common form of hernia is the inguinal, especially in young foals, where large inguinal rings are the prime cause (Wright, 1963). Spontaneous recovery normally occurs within 3–6 months and no long-term detrimental effects have been reported (Varner and Schumacher, 1991). Surgical intervention is sometimes required (Van der Veldon, 1988).

Apart from the mortal risk of intestinal strangulation, the biggest problem associated with testicular hernias is the effect on testicular function, due to elevated temperature from the close proximity of the intestine (Varner and Schumacher, 1991; Cox, 1993a, b). As discussed previously, an increase in testicular temperature has a direct effect on function (see Chapter 2).

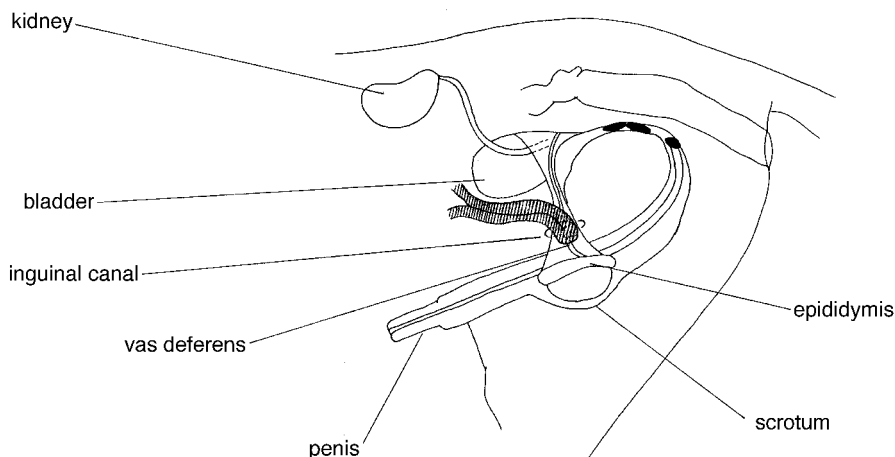


Fig. 19.4. An inguinal hernia in the stallion, in which a loop of the intestine folds through the inguinal ring.

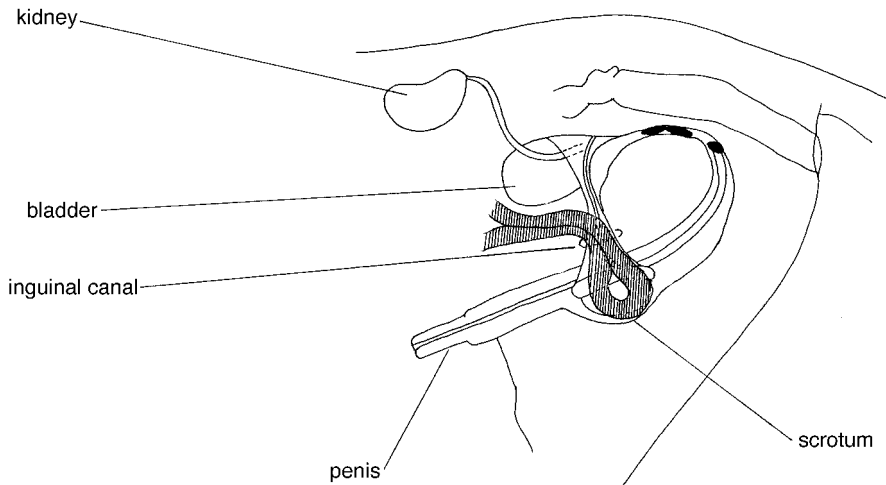


Fig. 19.5. A scrotal hernia in the stallion is a more extreme case of inguinal hernia, in which the loop of intestine has entered the scrotum and there is significant danger of complete ligation of the intestine, necrosis and death.

19.2.2.4.3. TESTICULAR HYPOPLASIA OR DEGENERATION. Both hypoplasia and degeneration are the terms given to an underdeveloped and therefore underfunctioning organ. Hypoplasia is generally the term given to a condition present from birth. Hence in the case of testicular hypoplasia, the testes, for some reason, have never developed beyond an immature stage (Roberts, 1986b; Varner and Schumacher, 1991; Varner *et al.*, 1991). Its causes are many, including cryptorchidism and hernias, but also malnutrition, endocrine malfunction, infections, irradiation and toxins and it may also be an inherited fault (Roberts, 1986b). The extent of the problem varies considerably from mild, where the testes appear normal, though possibly slightly small, to more severe cases, where the testes are significantly smaller than normal and, if the condition is advanced, the testes may have become hard due to the overdevelopment of connective tissue (Ladd, 1985). Spermatozoa production depends on the severity of the condition, varying from slight impairment to aspermic (no sperm at all). Any spermatozoa that are ejaculated have a higher incidence of abnormalities. In such cases, the libido of the stallion is often not affected (Varner and Schumacher, 1991; Varner *et al.*, 1991).

Testicular degeneration refers to the condition where testicular development did originally occur to some extent but some subsequent problem has resulted in a degeneration of the tissue. The testes are highly sensitive to extrinsic factors and so testicular degeneration is a major cause of infertility in the stallion. Unlike hypoplasia, degeneration is an acquired condition. Degeneration may be temporary or permanent; it may be unilateral (the cause being localized in origin) or bilateral (a systemic cause) (McEntee, 1970). The condition is evident as a shrinking of the testes, often showing small epididymides, with a reduced number of spermatozoa within (Watson *et al.*, 1994a). Spermatozoa counts are depressed (Roberts, 1986b) and a decline in spermatozoa output is observed, with an increase in the percentage of morphologically abnormal spermatozoa (Friedman *et al.*, 1991; Blanchard and Varner, 1993b). The causes of testicular degeneration are many and varied, the prime causes being elevated testicular temperature, scrotal haemorrhage, increased scrotal insulation due to scrotal oedema, scrotal dermatitis and cryptorchidism (McEntee, 1970; Varner and Schumacher, 1991; Blanchard and Varner, 1993b). Minor causes include toxins, tumours, obstructions of the vas deferens,

testicular torsion and old age (Rossdale and Ricketts, 1980; Pickett *et al.*, 1989; Varner and Schumacher, 1991; Varner *et al.*, 1991). Testicular degeneration is, in most cases, reversible, providing that the duration of the problem is limited and the causative problem can be alleviated. However, infective and traumatic degeneration is more likely to be permanent (Burns and Douglas, 1985; Blanchard and Varner, 1993b).

19.2.2.4.4. TESTICULAR TORSION. The extent to which the testes of a stallion may twist or turn and the resultant effect are variable. Torsion occurs most commonly in younger stallions. The twist may occur through an angle of up to 360°, a condition difficult to detect immediately, as the testes, on cursory examination, would appear to be positioned correctly. More commonly, the angle of twist is 180°, resulting in the epididymis being in a cranial position (towards the stallion's abdomen) (Hurtgen, 1987). A minor torsion may be transient and present just a little pain and a slight decrease in ejaculate spermatozoa concentration. Such torsions may correct themselves (Threlfall *et al.*, 1990). Major torsion can result in symptoms similar to those of orchitis (section 19.2.2.5.1) including acute colic pain, scrotal swelling and obstruction of the blood supply, which, if present in a chronic case, may lead to degeneration (Kenney, 1975a; Threlfall *et al.*, 1990). There is some dispute as to the effect of the condition on semen quality. It is evident that if degeneration does result then semen quality will suffer.

19.2.2.4.5. TESTICULAR TUMOURS. Testicular neoplasms are rare in horses, though their exact incidence rate is difficult to ascertain, as the majority of stallions are gelded at a young age. Neoplasms can largely be divided into two: germinal neoplasms (malignant germinal cells of seminiferous tubules) and non-germinal neoplasms (Bostock and Owen, 1975; Caron *et al.*, 1985; Morse and Whitmore, 1986; Schumacher and Varner, 1993). Such conditions normally occur in older stallions and cryptorchids (Vaillencourt *et al.*, 1979). Often, the condition is not associated with pain or elevated

temperatures, but a firm swelling may be felt in the testicular tissue and the testis affected is enlarged. Neoplasms are causes of testicular degeneration and therefore associated with depressed spermatozoa counts and a high incidence of morphological abnormalities (Hurtgen, 1987).

19.2.2.4.6. VAS DEFERENS AND ACCESSORY-GLAND PHYSICAL ABNORMALITIES. Physical abnormalities of the vas deferens and the accessory glands are rare but, if they occur, are invariably associated with a current or previous infection (section 19.2.2.5) or inherited abnormalities (Varner *et al.*, 1991). Abnormalities associated with infection are manifest as fibrous growths or swellings at inflammation sites, which may cause obstruction and aspermia. Congenital abnormalities may be evident as immature, underdeveloped structures or complete absence. Most abnormalities can be identified by rectal palpation or ultrasonic scanning.

19.2.2.4.7. PENIS AND PREPUCE PHYSICAL ABNORMALITIES. Abnormalities of the penis or prepuce are normally associated with trauma or injury. The penis, especially when erect, is very vulnerable to traumatic injury from a kick by an unreceptive mare (Vaughan, 1993). This causes vascular rupture and/or haemorrhage, making the return of the penis to within the prepuce difficult and painful. Haemorrhage of penile blood vessels may also occur if a stallion covers a mare with a Caslick's, prior to episiotomy, or if the mare suddenly lunges to one side while being covered. The long-term effects of such a trauma will depend not only on the physical recovery of the penis but also on the stallion's psychological recovery. Such a trauma can make a stallion, especially a young one, very reluctant to cover a mare again. Damage to the urethra within the penis is evident as blood contamination of semen, termed haemospermia (Schumacher *et al.*, 1995).

Tumours of the penis are not often malignant, but lesions due to melanomas, sarcomas, carcinomas and herpes virus often burst and haemorrhage at covering and

cause the stallion considerable pain. Blockage of the urethra or vas deferens has been reported, characterized by a normal libido but small-volume aspermatic semen, even though testicular function is normal.

19.2.2.5. Infectious infertility

19.2.2.5.1. TESTICULAR DISEASE AND INFECTION. Infection of the testes (orchitis) may have a systemic or localized cause. Either will result in elevated testicular temperature and associated decline in spermatogenesis. The magnitude of the decline in fertility rates and the time period reflect the severity of the disease and the duration of the problem. Recovery, when it occurs, will be somewhat delayed, as the prime site of effect, as far as spermatogenesis is concerned, is the germinal cells. The spermatogenic cycle being 56 days, this period of time must be allowed after recovery for semen quality to return to normal (Varner *et al.*, 1991).

Systemic disease, causing orchitis, normally results in bilateral inflammation of the testes and epididymis. Bacterial agents causing such orchitis include *Streptococcus equi* (strangles), *Streptococcus zooepidemicus*, *Klebsiella pneumoniae*, *Actinomyces bovis* and *Pseudomonas mallei* (glanders) and possibly *Salmonella abortus equi*. Viral agents may also be a systemic cause of orchitis: these include equine viral arteritis (EVA), equine infectious anaemia and equine influenza (Rossdale and Ricketts, 1980; Ladd, 1985; Roberts, 1986b; De Vries, 1993; Slusher, 1997). Systemic infections cause chronic, rather than acute, orchitis and have more of a chance of causing low-grade testicular degeneration and, with it, permanently depressed semen quality.

Localized infections may be caused via a wound, often to the scrotum, but may also be caused by infection via the inguinal canal (Varner and Schumacher, 1991). Such infections tend to cause acute orchitis (De Vries, 1993), which may be unilateral or bilateral and present initially as soft, flabby, swollen testes. If the condition persists, chronic orchitis may result. Semen quality will be poor, with a decline in spermatozoa concentrations and an increased incidence of abnormalities

(Hurtgen, 1987). The major infective agents associated with localized orchitis are *Staphylococcus* species, *Escherichia coli*, *S. zooepidemicus* and *S. equi*. In cases of acute orchitis, rises in testicular temperature are also a potential hazard (Blanchard and Varner, 1993b).

Orchitis may also be caused by trauma or parasites. The parasite most often associated is *Strongylus edentatus* larvae (Smith, 1973). These can migrate into the testicular tissue, causing orchitis or obstruction of the testicular artery within the pampiniform plexus. This will have an additional detrimental effect upon the efficiency of the counter-current heat-exchange mechanism (Roberts, 1986b; Varner *et al.*, 1993).

Finally, orchitis may be caused as a result of damage to Sertoli cells and hence the blood–testes barrier. This will result in an autoimmune response and inflammation (Papa *et al.*, 1990; Zhang *et al.*, 1990b).

19.2.2.5.2. VAS DEFERENS AND ACCESSORY-GLAND DISEASE AND INFECTION. Inflammation of the vas deferens and accessory glands is often associated with inflammation of the epididymis and is often the cause of epididymitis. Such inflammation is often accompanied by enlargement of the ampulla accessory gland, detected by rectal palpation. Seminal vesicles are also susceptible to infection (seminal vesiculitis) (Blanchard *et al.*, 1987). Infection by *Corynebacterium pyrogenes* and *Brucella abortus* is the most common cause of infection in these glands. Infection is often characterized by increased leucocyte concentrations within semen, especially in semen collected after rectal palpation. Rectal palpation will also reveal that the seminal vesicles are swollen and painful (Varner *et al.*, 1991).

19.2.2.5.3. PENIS, PREPUCE AND URETHRAL DISEASE AND INFECTIONS. Infection of both the penis and the urethra, both viral and bacterial, may occur, often resulting in haemospermia. Contamination of semen with blood not only indicates the risk of possible infection transfer but is also associated with low fertility rates. Infective agents causing haemospermia

include *Habronema* larvae (summer sores) (Hurtgen, 1987; Varner and Schumacher, 1991), *Strongylus edentatus* larvae (Pickett *et al.*, 1981), *Trypanosoma equiperdum* (dourine) (Couto and Hughes, 1993) and herpes virus (coital exanthema or genital horse pox).

Not only may the penis itself be infected, but it is also the major means by which venereal infection can be passed from the stallion to the mare and vice versa. The penis has a naturally balanced microflora, which causes no problem to the stallion or any mares that he covers. However, if this balance is disturbed, due to systemic infection or disease, general ill health or impaired normal disease resistance, serious consequences can result. Similarly, if he comes into contact with a contaminated mare, his natural microflora balance may be breached and this may allow the invasion of foreign infective agents. The prepuce area protecting the penis then provides an ideal environment in which such organisms can multiply. Contamination of the stallion's penis may result from poor hygiene, especially at covering and veterinary examination. Therefore, regular swabbing of the stallion and all mares to be covered and the washing of the stallion and mare during the preparation for covering go a long way to preventing venereal-disease transfer.

The stallion is often asymptomatic, failing to demonstrate any clinical signs of infection, but infection may be traced back through symptoms shown by mares he has covered. Details of the organisms involved and their potential effect on reproductive performance are given in the following section dealing with mare infertility (section 19.3.2.5), as it is in the mare that these infections manifest their symptoms. Isolation and treatment of the stallion are the only course of action when such infections are suspected. Transfer from mare to mare via a stallion in a busy season is very easy and can have disastrous consequences.

Other conditions or infections may be transferred via the stallion at covering. For example, coital exanthema, often, but not always, characterized by lesions, particularly in the warmer climates of Asia, Africa, South

America and South-East Europe (Pascoe and Bagust, 1975; Couto and Hughes, 1993). EVA (Timoney *et al.*, 1988), equine herpesvirus (EHV3 and possibly EHV1) (Pascoe and Bagust, 1975; Jacob *et al.*, 1988), habronemiasis lesions (summer sores) (Philpott, 1993) and fungal infections (Zafracas, 1975) are other examples.

Complete prevention of venereal disease is difficult, but is aided by adhering to full hygiene precautions prior to covering, though complete disinfection of the stallion's penis is impossible and not advisable. Regular swabbing in accordance with the Horse Race Betting Levy Board guidelines (Horse Race Betting Levy Board, 2001) will greatly increase the chance that any bacteria present are identified so that appropriate treatment can be given. Treatment itself can cause problems, as systemic antibiotics may affect the natural microfloral balance, which will take time to restore. Topical application to such a sensitive area may also cause dryness and cracking, causing pain at covering.

19.2.2.6. Immunological infertility

Semen contains many antigens, including those within seminal plasma and those that are spermatozoa-bound. Under certain conditions, an autoimmune response to these antigens may occur, causing the destruction of spermatozoa within both the testis and the female tract (Wright, 1980; Teuscher *et al.*, 1994).

19.2.2.7. Semen abnormalities

Semen abnormalities are discussed in full in Chapter 20, along with the evaluation of semen. In summary and as indicated throughout the previous text, most infections and trauma of the male reproductive tract have an adverse effect on sperm production and hence fertility. The normal parameters expected of a semen sample are given in Table 20.3.

Infection and/or abnormalities of the reproductive tract may affect any of these parameters but usually cause a reduction in

sperm concentrations, inadequate motility and poor longevity. Infection, rather than trauma, is often characterized by high leucocyte counts. If a semen sample does not meet the required parameters, the cause of the problem should be identified before the stallion is used, for his own protection and that of any mare. In addition to poor sperm quality, urospermia (urine contamination) and haemospermia (blood contamination) may be evident (Voss and McKinnon, 1993).

Haemospermia, as mentioned previously, may be caused by a number of infective agents or by physical damage (Schumacher *et al.*, 1995). A red blood-cell count $> 500 \text{ ml}^{-1}$ or a white blood-cell count $> 1500 \text{ ml}^{-1}$ will have an effect on fertility. This effect appears to be modulated by the white or red blood cells, rather than via the serum (McKinnon *et al.*, 1988b).

Urospermia has a number of causes, including neurological disorders (Rasbech, 1975; Leendertse *et al.*, 1990; Mayhew, 1990; Samper, 1995a; Lowe, 2001). Stallions suffering from urospermia may appear normal, with no neurological defects and with adequate libido and mating ability. The condition may be continuous, intermittent and unpredictable. Contamination may occur at any time during ejaculation and may be as little as 1 ml or as great as 250 ml (Varner and Schumacher, 1991). Evidence suggests that contamination is not likely to be due to just leakage but to an all-or-nothing effect (Nash *et al.*, 1980). Urine contamination within a semen sample adversely affects spermatozoa motility and their capacity to fertilize an ovum (Hurtgen, 1987; Varner and Schumacher, 1991). Urine contamination may also affect semen pH (Hurtgen, 1987; Samper, 1995a). The severity of the problem is dose-dependent and stallion spermatozoa can tolerate minute amounts of urine without deterioration.

19.3. Mare Infertility

On average 40–80% of services result in a live foal. This figure, however, varies signifi-

cantly with the population of mares (Osborne, 1975; Sullivan *et al.*, 1975; Mahon and Cunningham, 1982; Baker *et al.*, 1993). Human interference in the reproduction of the horse has also had a significant effect on reproductive performance. Live foal rates for wild horses and ponies are in the region of 95%, compared with 60% for hand-bred mares (Bristol, 1987). Embryo mortality or early embryonic death (EED) is held responsible for a significant amount of the apparent infertility. Rates of 5–14% have been reported for in-hand mating, compared with 1–2.5% for free mating, though higher rates of EED (up to 50% for normal mares) have been reported by others (Baker *et al.*, 1993). As most EED occurs prior to day 40 (Ginther, 1985), most measurements of infertility will also include this.

Reproductive performance has been largely ignored in the improvement of the equine. This is in contrast with other farm livestock, where reproductive efficiency is of utmost importance. It is evident that barrenness in a mare at the end of the season is not necessarily due to pathological infertility but is also a consequence of other environmental and managerial influences and the natural tendency in many mares to take a season off breeding. Some mares are consistently barren in early life but successfully breed later on. Failure to produce an offspring in a particular year is therefore a complicated problem and is not to be confused with infertility and EED. Before the reasons for a mare failing to produce a foal are discussed in detail, the following glossary should be noted to prevent confusion in terms.

Fertile	able to produce a live foal
Infertility	a temporary inability to reproduce
Barrenness	lack of a pregnancy at the end of the season, but perfectly capable of producing a foal, as demonstrated in previous years
Subfertility	inability to reproduce at full potential, may be temporary or permanent
Sterility	a permanent inability to reproduce

Embryo mortality or death (EED)	embryo loss prior to day 40, often occurring between scanning at day 15 and day 40
Abortion	fetal death after day 40
Stillborn	fetal death after day 300
Fertilization rate	number of ova fertilized per ovulation (85–90%) (Pycock, 2000)
Pregnancy rate	number of mares pregnant on a specified day, expressed per oestrous cycle (45–60%) or per breeding season (75–90%) (Pycock, 2000)
Live-foal rate	number of mares foaling per number of mares bred over the season (50–85%) (Pycock, 2000). Arguably the best indicator of reproductive performance

In the following account, the term reproductive performance will be used, as it encompasses all the above definitions. In many texts, the strict definitions of infertility, EED and abortion are not adhered to and this makes it very hard to distinguish precisely what is responsible for reported changes in reproductive performance. In the mare, reproductive performance does not just depend upon successful gamete production, as is the case with the stallion, but also upon an appropriate environment for fertilization, free-living embryo, implantation, placentation and subsequent parturition. As such, the following sections will include consideration of fertilization failure, EED and abortion, as appropriate. The failure of a mare to produce a foal at the end of a season may have numerous causes and, as in the case of the stallion, can be divided into extrinsic and intrinsic factors.

19.3.1. Extrinsic factors affecting reproductive performance in the mare

Extrinsic factors affecting reproductive performance in the mare may be considered to include: lack of use; subfertile or infertile stallion; poor stallion management; poor

mare management; and the artificially imposed breeding season.

19.3.1.1. Lack of use

A mare may not be covered in a particular season due to design or unavoidable circumstances. If she foals late in one season, she may not be re-covered, in order to allow her to return to foaling earlier in the year. Disease or infection may preclude a mare from use in a particular year, either because she herself is not in a fit condition to successfully carry a foal or there is a danger of systemic- or venereal-disease transfer. In the case of a performance horse, she may not be bred in a particular year due to work commitments, though advances in embryo transfer now potentially allow such mares to foal via use of a recipient mare.

19.3.1.2. Subfertile/infertile stallion

Half the responsibility for the success or failure of a covering lies with the stallion and half with the mare. If a mare is covered by an infertile or subfertile stallion, her chances of producing a foal are significantly reduced through no fault of her own. The causes of infertility in the stallion have already been discussed in the previous section. Mares can only be expected to perform to their full reproductive potential if they are covered by a stallion whose semen meets minimum requirements. A stallion must also be physically capable of covering a mare effectively; a good semen evaluation in the absence of the ability or willingness to cover is of no use in the natural service of a mare. To a certain extent, this problem may be overcome by the use of AI, but in such cases it must be certain that the stallion's lack of libido or ability is not due to a potentially heritable fault.

19.3.1.3. Poor stallion management

Stallion management has been considered in detail in Chapters 12, 13 and 18 and earlier in this chapter. It is evident that, if any aspect of a stallion's management, especially his cov-

ering management, is not correct, then there is the potential for his fertility rates to be affected. Management in the earlier formative years also has a significant effect on a stallion's libido and hence reproductive performance.

The imposition of an artificial breeding season causes problems with reproductive performance. Even with the use of artificial lights, a stallion's performance during the months of December to February is lower than during his true breeding season, so fertility rates for mares covered within this period of time cannot be expected to meet normal expectations.

Behavioural abnormalities, often associated with inappropriate management, such as failure to obtain or maintain an erection, incomplete intromission and ejaculation failure, may cause apparently low fertilization rates, as can the incorrect detection of successful ejaculation (Kenney, 1975a; Pickett and Voss, 1975b).

19.3.1.4. *Poor mare management*

Mare management has been discussed in detail in Chapters 12, 13 and 14. Any deficiencies or inadequacies in brood-mare management can lead to poor reproductive performance. Of specific significance is covering management, especially oestrus detection. Better, more experienced management and the use of veterinary diagnosis and hormonal manipulation of the cycle are seen to significantly improve reproductive performance. Stress associated with handling and transportation is suggested to be associated with EED (Osborne, 1975), possibly via changes in plasma cortisol (Bacus *et al.*, 1990) and progesterone concentrations (Van Niekerk and Morgenthall, 1982). Finally, nutritional stress is also reported to affect reproductive success, again mainly via EED and abortion (Henneke *et al.*, 1984; Potter *et al.*, 1987; Ball, 1993a).

19.3.1.5. *Imposed breeding season*

As detailed previously and in Chapter 3, there is currently considerable pressure for

foals to be born as soon as possible after 1 January. The methods by which the mare's breeding season may be manipulated in order to achieve this are discussed in Chapter 12. Regardless of the treatment used, pregnancy rates out of the natural breeding season are never as high as normal expectations. As such, the continued imposition of an arbitrary breeding season places unfair constraints upon a mare's potential reproductive performance.

19.3.2. **Intrinsic factors affecting reproductive performance in the mare**

Intrinsic factors affecting reproductive performance in the mare may include age, chromosomal, hormonal, pituitary, ovarian, Fallopian tube, uterine, cervical, vaginal and vulval abnormalities and infections. All these will be discussed in turn; many, however, are closely interrelated.

19.3.2.1. *Age*

Age is reported to have the most significant bearing on reproductive performance (McDowell *et al.*, 1988; Barbacini *et al.*, 1999). In general, fertility declines with age and EED increases (Ball *et al.*, 1989; Ball, 1993a; Carnevale *et al.*, 1993). Baker *et al.* (1993) indicated that young mares have 90–95% fertilization rates, with EED up to 50%. However, the fertilization rates of older mares decline to 85–90% and EED may reach 100%. Work investigating the viability of embryos indicates that significantly more cleaved ova are collected at day 15 in 2–10-year-old mares, compared with 20-year-old mares. Additionally, older mares yield more embryos with morphological abnormalities (Carnevale and Ginther, 1992; Carnevale *et al.*, 1993) and demonstrate multiple ovulations and so an increased likelihood of multiple pregnancies (Davies Morel and O'Sullivan, 2001), thus further adding to the high apparent infertility rates in older mares. Evidence would also suggest that the occurrence of anovulatory oestrus is greater in mares

over 20 years (Vanderwall *et al.*, 1993). Despite reducing reproductive performance, providing a mare is in good physical condition, she may breed successfully well into her twenties. However, welfare considerations may preclude breeding a mare of advanced age. In such cases, embryo transfer may prove a viable alternative to putting an older mare through the stresses of carrying a pregnancy to term (Carnevale and Ginther, 1992).

19.3.2.2. Chromosomal abnormalities

The normal chromosomal complement for the equine is 64 (32 pairs), the female complement being denoted as 64XX. Various variations on the normal complement include 63XO (a female with a single X chromosome), termed Turner's syndrome, one of the more common chromosomal abnormalities. Such individuals are characterized by small rudimentary ovaries, flaccid, poorly developed uterus, no ovarian activity and therefore permanent anoestrus. Such mares tend also to be short in stature (Chandley *et al.*, 1975; Hughes *et al.*, 1975). Mosaic chromosomal configuration may occur as a 64XX complement in some cells and 63XO in others; such individuals demonstrate erratic oestrous cycles with no ovulation (Chandley *et al.*, 1975). Very rarely, chromosomal complements of 64XX/65XXY are found; such horses are termed intersex (Dunn *et al.*, 1981; Halnan, 1985). Positive diagnosis of chromosomal abnormalities is only possible by genetic mapping via cytogenetic analysis of blood samples, though they may be indirectly indicated physiologically (Ricketts, 1975b, 1978; Halnan, 1985; Bowling *et al.*, 1987; Bowling and Hughes, 1993).

19.3.2.3. Hormonal abnormalities

As discussed in Chapter 3, the control of the mare's reproduction is a finely balanced cascade and interrelationship of hormones, involving the hypothalamic-pituitary-ovarian axis. Abnormalities/inefficiencies in any of these centres can cause imbalance

throughout the whole axis. The majority of hormonal deficiencies are associated with pituitary abnormalities – for example, Cushing's syndrome (Pycock, 2000). Complete failure, or neoplasia, of the pituitary is relatively rare in the horse, but temporary malfunction may occur, especially in association with the transitional period at the beginning or end of the breeding season. Mares during this transitional period tend to suffer from delayed oestrus, prolonged oestrus and dioestrus, silent ovulations, split oestrus (oestrus over a period of up to 3 weeks with possibly a quiescent period in the middle), etc. In such cases, it is evident that a period of time is required to allow the mare's system to re-establish regular 21-day cycles.

Diagnosis of hormonal abnormality is initially via a mare's behaviour and the seeming inability to detect oestrus or, conversely, apparent continual oestrus. Diagnosis of the cause is helped via scanning and rectal palpation, by which ovarian activity can be monitored. The incidence of hormonal deficiencies or abnormalities is particularly evident today, as we continually attempt to breed mares earlier in the season. As discussed in Chapter 12, the use of exogenous hormonal treatments and/or light treatment does successfully advance ovulation and oestrus within the year, but does not eliminate the transition period, which may still be associated with problems.

Pituitary or hypothalamic tumours are rare in mares; they are associated with muscle wasting, hypoglycaemia, docility, alopecia, blindness and uncoordinated movement, in addition to prolonged anoestrus.

Hormonal deficiencies during pregnancy may also result in reproductive failure. In particular, progesterone insufficiency may result in EED or abortion, depending upon when it occurs (Ginther, 1985; Morgenthal and Van Niekerk, 1991). Exogenous progesterone supplementation has proved successful with many mares, and it is routinely used in some stud practices, though such use is not normally justified with normal mares (Ganjam *et al.*, 1975; Pycock, 2000).

19.3.2.4. Physical abnormalities

19.3.2.4.1. OVARIAN ABNORMALITIES. Occasionally, ovaries may be absent, due to surgical intervention or chromosomal abnormality. Inactive ovaries and ovulation failure are often observed in mares and are exacerbated by the imposition of an arbitrary breeding season (Kenney *et al.*, 1979).

19.3.2.4.1.1. Follicular atresia. Follicular atresia is responsible for some incidences of ovulation failure. In such cases, a group of follicles will develop normally to about 3 cm in diameter, but there is a failure in the emergence of a dominant follicle, which would normally develop further. Conditions such as ovarian hypoplasia, granulosa cell tumours, ovarian cysts, uterine infections and malnutrition have all been implicated in follicular atresia (Bosu, 1982; Pugh, 1985; Bosu and Smith, 1993). The best cure appears to be time, especially in mares encountering problems during the transitional stage of the breeding season. Often, succeeding cycles will not demonstrate the condition.

19.3.2.4.1.2. Corpora lutea persistence and failure. Corpora lutea (CL) persistence and conversely failure are also causes of reproductive failure in the mare, manifesting themselves as long or short oestrous cycles, respectively. Failure of the CL is less evident in the mare than persistence. However, CL failure is implicated in experiments using progesterone supplementation to prevent abortion (Ganjam *et al.*, 1975; Allen, 1993). The effect of progesterone insufficiency has also been considered in section 19.3.2.3. The presence of a persistent CL is more common in mares and is an important cause of anoestrus. The normal lifespan of a CL is 14 days, after which, in the absence of a pregnancy, the luteolytic hormone prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), secreted by the uterine endometrium, takes effect. A persistent CL is presumably, therefore, a result of failure in the release of $PGF_{2\alpha}$ or in the ability of the CL to react appropriately. The presence of such conditions in a mare is implicated by the lack of oestrous behaviour, and is confirmed by scanning or rectal palpation. The failure of the

luteolytic message may be linked to uterine infection, rendering the uterine endometrium unable to produce $PGF_{2\alpha}$. EED and associated pseudopregnancy are other potential causes of a persistent CL. Treatment with exogenous $PGF_{2\alpha}$ is normally successful (Kenney *et al.*, 1975b; Pycock, 2000).

19.3.2.4.1.3. Anovulatory follicles. Anovulatory follicles (ovarian cysts or luteinized unruptured follicles) can be a cause of anoestrus. Most cysts are associated with follicles, though their presence must not be confused with large, perfectly normal follicles, which have been reported to reach up to 10 cm in diameter. Anovulatory follicles are characterized as large follicles that fail to rupture and ovulate. Instead, over time, they fill with blood and persist as haematomas, possibly over a number of cycles. Their presence is further complicated by their secretion of progesterone – hence their alternative name of luteinized unruptured follicles and their similarity to functional CL. Differentiation between the two can only be made at scanning by variations in their echogenic characteristics (Ginther and Pierson, 1989; Pycock, 2000).

19.3.2.4.1.4. Ova fossa cysts. Ova fossa cysts are reported, especially in older mares. They appear to be associated with the epithelium of the fimbriae and may cause blockage of the ova fossa and therefore a disruption of ova release and fertilization. They are often evident as a bundle of cysts, similar to a bunch of grapes, near the ova fossa. In the extreme, they may also interfere with the blood supply to the rectum (Prickett, 1966; Stabenfeldt, 1979).

19.3.2.4.1.5. Granulosa theca cell tumours. Granulosa theca cell tumours are the most common tumours within the equine ovary and an important cause of anoestrus (Sundberg *et al.*, 1977). They normally affect mares between the ages of 5 and 7 and are usually associated with a single ovary. The ovaries are either polycystic or large solid structures and may weigh up to 8 kg (Norris *et al.*, 1968; Fig. 19.6).



Fig. 19.6. Granulosa theca-cell tumour in the mare, evident in the polycystic form.

The symptoms demonstrated by such mares depend on the hormones secreted by the tumours. Oestrogen, the most common, may result in nymphomaniac behaviour (prolonged oestrus) in the absence of ovulation. Testosterone-producing cysts result in stallion-like behaviour. Elevated inhibin concentrations are thought to cause the characteristically small contralateral ovary, due to negative-feedback effects (Piquette *et al.*, 1990). Removal of the affected ovary allows the resumption of normal reproductive activity by the remaining ovary (Meager, 1978).

19.3.2.4.1.6. Ovarian teratomas. Ovarian teratomas, arising from germ cells and containing hair, teeth, bone, cysts, etc., have been reported but very rarely occur (Rossdale and Ricketts, 1980; Hughes, 1993) and, unlike teratomas seen in stallions, are benign. They are unilateral in occurrence, allowing the other ovary to function normally; pregnancy rates may or may not be affected significantly. Parovarian or paroophoron cysts are also reported, but rarely cause problems.

19.3.2.4.1.7. Hypoplasia. Ovarian hypoplasia (underdevelopment) is a further cause of anoestrus. It is characterized by small, immature ovaries, with no ovarian activity or increase in ovarian size within the breeding season. Hypoplasia is usually bilateral

and is often associated with chromosomal or hormonal abnormalities, as previously discussed.

19.3.2.4.1.8. Cystic ovaries. The term cystic ovary implies the presence of fluid-filled structures within the ovarian stroma, which are hormonally active. Such structures are reported in cattle but are reported not to occur in mares (Pycock, 2000).

19.3.2.4.1.9. Other abnormalities. Dysgerminomas (malignant tumours of germ-cell origin), abscesses and haematomas (overflowing of the follicular cavity with blood after ovulation) are also reported to occur, but rarely (Meuten and Rendano, 1978; Bosu *et al.*, 1982; Neely, 1983; Bosu and Smith, 1993).

19.3.2.4.1.10. Multiple ovulations. Multiple ovulations, and the resulting multiple pregnancies, have been considered previously in Chapters 3 and 14. However, they warrant mentioning here as a major cause of EED and abortion and hence reproductive failure. The mare's tract is not competent to satisfactorily maintain more than one fetus. Fetal restriction results, causing either spontaneous abortion of one or both fetuses or the mummification of the smaller twin (McDowell *et al.*, 1988). Twins are therefore not desirable and management in general is

geared towards avoiding them. Twins detected by scanning in early pregnancy are routinely aborted (using PGF_{2α}) so that the mare is in essence infertile to that covering, or one twin is 'pinched out', potentially allowing the remaining embryo to go to term, but with an increased risk of EED or abortion. Twinning is an inherited trait and can be avoided or an awareness of its possibility gained by studying a mare's breeding history and any previous incidents of twinning (Jeffcote and Whitwell, 1973; Ginther, 1982; Miller and Woods, 1988).

19.3.2.4.2. FOLLICULAR-TUBE ABNORMALITIES. The extent of follicular-tube abnormality is disputed. Arthur (1958), using slaughterhouse material, demonstrated that the incidence of such abnormalities was very low. However, Vandeplasse and Henry (1977) demonstrated a 40% incidence, normally involving adhesions of the infundibulum to other parts of the reproductive tract. Collagenous masses, which may occlude the lumen of the follicular tubule, have been more recently documented (Lui *et al.*, 1990). Rarely, an ovarian cyst may be seen to block the entry to the Fallopian tube at the infundibulum. Tumours of the Fallopian tube are extremely rare (Allen, 1979).

19.3.2.4.3. UTERINE ABNORMALITIES. Uterine abnormalities due to congenital defects or infections are relatively well understood in the mare, compared with abnormalities of the remainder of the tract. The development of techniques such as ultrasonic scanning, endoscopy and uterine biopsy has significantly advanced our understanding of uterine physiology and pathology (Kenney, 1975b; Brook, 1993).

19.3.2.4.3.1. Hypoplasia. Uterine hypoplasia (underdevelopment) is characterized by an inability to develop adequately in order to maintain a pregnancy. The endometrial glands are those most significantly affected, tending to be very small and so incapable of adaptation to support a pregnancy. As a result, even if fertilization does occur, the EED rate is high. Covering mares

too close to puberty is associated with high rates of EED, due to hypoplasia, simply because the uterine development to date is inadequate. The actual age at which the uterus is fully mature depends very largely on the individual mare, 18 months to 4 years is considered acceptable. Hypoplasia at 4 years old or over is indicative of a problem, which is likely to be permanent and may be associated with chromosomal or hormonal abnormalities (Ricketts, 1975a, b).

19.3.2.4.3.2. Hyperplasia. Uterine hyperplasia (overdevelopment) is characterized by an overdevelopment of the uterus for the reproductive stage of the mare or a failure to recover from a previous event, such as pregnancy. Again, the endometrial glands are most significantly affected. As indicated, the condition is often a result of delayed involution post-partum, the uterine endometrium failing to return to normal within the time expected (Ricketts, 1975a; Kenney, 1978; McKinnon, 1987c; McKinnon *et al.*, 1988a). Hyperplasia may also be a result of EED or abortion or a consequence of hormonal imbalance, often resulting from hormone-secreting tumours. Hyperplasia is normally a temporary condition, which can be reversed by reproductive rest or hormonal treatment (Bosu *et al.*, 1982; Van Camp, 1993).

19.3.2.4.3.3. Uterine atrophy. Mares with uterine atrophy or senility are normally characterized as repeat breeders with high rates of EED (Greenhof and Kenney, 1975; Kenney, 1978; Van Camp, 1993). Uterine atrophy is caused by a decrease in the number of endometrial glands, due to atrophy or an inability to regenerate themselves. It is often associated with chromosomal intersex conditions, ovarian incompetence or progressive wear and tear in multiparous mares. It is also reported to have a greater occurrence late in the breeding season, presumably due to a decline in oestrous and ovarian activity. Generally, such late-season atrophy is of little concern, but evidence of it occurring early in the season may be indicative of a permanent problem and effect on reproductive performance. This condition is normally irreversible.

19.3.2.4.3.4. Uterine fibrosis. Uterine fibrosis is a degenerative uterine change, most commonly found in old multiparous mares; it is characterized by fibrotic changes around the endometrial glands, forming glandular nests. As a result, the secretions of the endometrial glands decrease and the glands dilate, increasing the incidence of uterine cysts and resulting in increasing EED, due to a disruption of embryo mobility, or abortion in late pregnancy, due to restricted placental size (Van Camp, 1993).

19.3.2.4.3.5. Uterine luminal cysts. Uterine luminal or endometrial cysts are the most common form of uterine lesion (Eilts *et al.*, 1995). They are generally thin-walled, greater than 3 cm in diameter and filled with lymph and may occur singly or in multiples (Fig. 19.7). They are particularly evident in mares of 10 years old or over. Their effect on reproductive performance is disputed. If they are present in any number, they are likely to interfere with embryonic mobility, increasing EED (McDowell *et al.*, 1988), and also to reduce the uterine surface area available for placental attachment, increasing abortion rates (Curnow, 1991). Treatment may be attempted by puncturing the cysts

via curettage, endoscopic manipulation or thermocautery, though they may subsequently recur (Mather *et al.*, 1979; Neely, 1983; Wilson, 1985; McDowell *et al.*, 1988; Pycock, 2000).

19.3.2.4.3.5.1. Uterine curettage. Uterine curettage was traditionally used as a treatment for a whole range of conditions that resulted in damage to the uterine endometrium. It works on the principle of mechanical or chemical irritation of the endometrium, the rationale being that irritation stimulates and initiates a cleansing and regeneration process within the uterine endometrium and a mobilization of neutrophils to the affected site. Mechanical curettage involves physically scraping the entire surface of the endometrium, using a cutting edge mounted upon a long shaft that is passed through the cervix into the uterus. Chemical curettage involves the infusion of a chemical irritant, e.g. povidone-iodine or kerosene, which is reported to have a similar effect (Bracher, 1992). Curettage was once very popular but has been largely discredited as ineffective, with the potential to cause excessive scar tissue, haemorrhage and uterine adhesions.

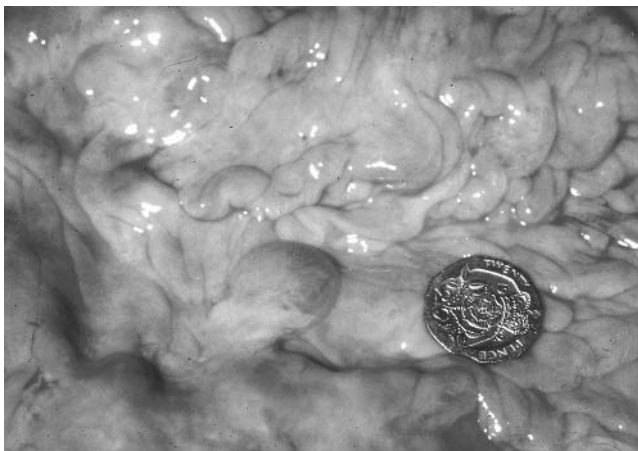


Fig. 19.7. A single luminal cyst within the uterus. They may also be present in multiples. Luminal cysts do not normally cause excessive problems unless evident in large numbers, where they may interfere with embryo mobility and subsequent implantation.

19.3.2.4.3.6. Ventral uterine dilatation.

Ventral uterine dilatation or sacculation is caused by uterine myometrial atrophy, normally in the base of one uterine horn, forming an outfolding or sacculation, which often collects fluid. This is again more common in older multiparous mares, due to a weakening of the myometrium. It often occurs at the implantation site, and may be caused by a gradual weakening of the wall in an area of repeated excessive stretching. Treatment is, unfortunately, relatively unsuccessful, but some beneficial results have been reported using oxytocin, or oxytocin in combination with warm saline lavage (Kenney and Ganjam, 1975; Neely, 1983). The fluid accumulation is normally of greatest concern, making such mares susceptible to chronic endometritis and pyometra (Kenney and Ganjam, 1975; Brinsko *et al.*, 1990).

19.3.2.4.3.7. Uterine adhesions.

Uterine adhesions are present as single or multiple bands or sheets of tissue within or across the lumen of the uterus and are the result of uterine trauma from dystocia, intrauterine infusion or severe endometritis or after treatment with caustic solutions. Their effect upon fertility depends upon their extent, but they may disrupt embryo mobility, restrict placental attachment or even cause post-partum problems, such as placental and fluid retention, leading to endometritis. As such, they may be associated with EED or abortion. Attempts may be made to remove or break adhesions manually via an endoscope and biopsy forceps or electrocautery (McKinnon, 1987a, b; Van Camp, 1993).

19.3.2.4.3.8. Uterine neoplasia.

Neoplasia or tumours within the uterus are very rare, but when present can be evident as single or multiple nodules and cause persistent haemorrhage. Treatment may be attempted by surgery or endoscopy with some success (Bostock and Owen, 1975; Kenney, 1978).

19.3.2.4.3.9. Foreign bodies.

Very occasionally, foreign bodies, such as fetal bone, tips of uterine swabs, frozen semen straws, etc., are found within the uterus, resulting in

chronic endometritis. Their removal, followed by treatment and recovery time, normally restores reproductive performance (Ginther and Pierson, 1984c; Pycock, 2000).

19.3.2.4.4. CERVICAL ABNORMALITIES. Cervical abnormalities normally arise from damage at parturition. Lacerations or injuries to the cervix often do not heal properly. They can cause adhesions, which may block the entrance to the uterus through the cervix or cause cervical incompetence. This will inhibit sperm deposition and allow infection to enter the uterus (Sertich, 1993). As discussed in some detail in Chapter 1, the cervix naturally forms the final seal protecting the upper reproductive tract from infection. Minor adhesions may be treated by physically cutting or electrocauterizing the scar tissue and inserting a plastic tube to prevent reoccurrence, and lacerations can be surgically corrected (Brown *et al.*, 1984; Aanes, 1993). The prognosis in most cases, however, is not good. Neoplasms of the cervix are very rare (Sertich, 1993). Inherited cervical incompetence, though, has been reported in pony mares (Lieux, 1972; Brown, 1984).

19.3.2.4.5. VAGINAL ABNORMALITIES. Vaginal abnormalities have several causes. Among these is damage at parturition, often a result of fetal malpresentations. Superficial damage will correct and heal naturally, though there is the risk of adhesions. Severe adhesions may cause the mare pain at subsequent coverings. In the extreme, rectal-vaginal fissures may be opened up by the foal's foot passing through the top of the vagina and into the rectum during parturition. The prognosis in such cases depends on the length of the opening formed, but can be very poor when substantial rectovaginal fissures occur (Spensley and Markel, 1993; see Chapter 15). Occasionally, a persistent hymen may be evident. The hymen divides the anterior and posterior vagina and, if not broken prior to the first service, may tear, causing the development of scar tissue. A persistent hymen may also impede natural drainage and lead to pyometra.

Two of the most common vaginal abnormalities are associated with poor perineal conformation, pneumovagina and urinovagina. Pneumovagina (inspiration of air and bacteria into the vagina) is a common cause of infertility, especially in Thoroughbred mares, and is due to the incompetence of the vestibular and vulval seals and associated poor perineal conformation (Chapter 1). This can be alleviated quite successfully by a Caslick's vulvoplasty operation. Full details of vulval incompetence, its consequences and its treatment have already been given in Chapter 1.

Urinovagina (urine pooling within the vagina) results from weakness within the vaginal walls and a collection of urine within the resultant sacculaton. It is often associated with infection, which can easily spread to the rest of the reproductive tract and hence adversely affect reproductive performance. The condition is normally observed in older multiparous mares, with pendulous reproductive tracts, due to continual stretching and weakening with successive pregnancies. Occasionally, it may also be seen as a temporary phenomenon at foal heat, but in most circumstances it will have rectified itself by the second heat postpartum. If it is evident, it is essential that the mare is not covered, as there is an increased chance of *post coitum* endometritis. Treatment using oxytocin has proved reasonably successful (Monin, 1972; Brown *et al.*, 1978).

19.3.2.4.6. VULVAL ABNORMALITIES. Vulval abnormalities most commonly involve inappropriate perineal conformation, as discussed in detail in Chapter 1. The resulting vulval-seal incompetence increases the chance of infection entering the reproductive tract. Lacerations of the vulva occurring at parturition or due to accidental injury can again compromise vulval-seal competence, due to incorrect healing and adhesion formation. Failure to cut a Caslick's vulvoplasty prior to covering or parturition will also cause tearing and will predispose to adhesion formation and inappropriate vulval healing (Aanes, 1969, 1973, 1993).

Haemorrhage of the vulval lips may be evident, caused by the bursting of varicose veins. This has a minimal direct effect on reproductive ability but may cause discomfort at breeding. Neoplasms of the vulva are reported, most commonly melanomas, originating in the pigment-producing cells of the skin, especially prevalent in grey mares. These tumours can spread from the perineal area around the anus and eventually throughout the whole of the body. Squamous-cell carcinoma, normally associated with the penis, may also be seen on the vulval lips. Finally, enlarged clitorises, sometimes in the form of a vestigial penis, may be observed and are associated with chromosomal abnormalities and such animals are sterile.

19.3.2.5. Infectious infertility

19.3.2.5.1. OVARIAN INFECTIONS. As far as infection or disease is concerned, the ovary is essentially unaffected and the vast majority of ovarian abnormalities, and hence ovarian infertility, are not the result of pathogenic agents.

19.3.2.5.2. FOLLICULAR-TUBE INFECTIONS (SALPINGITIS). Salpingitis, inflammation of the Fallopian tubes or salpinges, is rarely seen, however, it may occur as a consequence of endometritis. Complete blockage of the Fallopian tubes is rare, but inflammation can interrupt the process of fertilization, the passage of ova towards the utero-tubular junction and sperm movement towards the ampulla. Infertility or subfertility may result. Occasionally, infection may cause inflammation of the valve at the utero-tubular junction, affecting the passage of sperm and/or fertilized ova.

19.3.2.5.3. UTERINE INFECTIONS. One of the major causes of infertility in the mare is endometritis (Bennett, 1987). Endometritis, inflammation of the uterine endometrium, is primarily caused by infection by venereal and opportunistic bacteria. Less commonly, non-infectious degenerative endometritis may occur. The main consequence of endometritis is a uterine environment hostile to embryo survival and implantation, result-

ing in EED and abortion. Endometritis is evident in four forms: acute endometritis; chronic endometritis; acute metritis; and pyometra. These will be discussed in turn later in the chapter.

There are several factors that predispose the mare's tract to infections, including immunological, physiological or endocrinal deficiencies, which may be inherited, leading to a predisposition to endometritis.

Unfortunately, the mare's reproductive tract is not well designed for the easy removal of infective organisms or the resulting exudate. Infections are difficult, therefore, for the mare's system to eliminate naturally and can easily develop into chronic infections. Chronic infections that are not identified can cause serious problems to the mare if not treated in good time. Temporary infertility is nearly always evident with endometritis and, if the infection damage is great, permanent reduction in reproductive performance will result. Bacterial infection is nearly always introduced at covering or by inadequate hygiene precautions during internal examination or immediately post-partum. It is therefore most important that, during covering and manipulation or examination of the mare's tract, strict hygiene precautions are adhered to (see Chapter 13).

One of the major problems with uterine infections is that they may remain undetected for prolonged periods of time, thus not only reducing the mare's reproductive performance, but also risking transfer to the stallion and hence to other mares. Regular swabbing is not only compulsory in some studs, but is good practice in order to ensure that all chronic endometritis infections and latent asymptomatic infections are identified and treated immediately.

Endometritis is often characterized by excess mucus, which may be seen exuding from the vulva, high leucocyte counts and increased uterine blood flow. Oedema (fluid accumulation) can be identified by scanning and the uterus can be felt, via rectal palpation, to be large and flaccid. The mare may also show shortened oestrous cycles, due to the irritation of the uterine wall resulting in premature CL regression.

19.3.2.5.3.1. Potential endometritis-causing bacteria. As indicated, endometritis is primarily caused by bacterial infection. There are six major bacteria that cause endometritis, with up to 15 different bacteria identified in some cases (Pycock, 2000). The six major bacteria will be considered and can be classified as opportunistic or venereal.

19.3.2.5.3.1.1. Opportunistic bacteria. Opportunistic bacteria are those that are common within the environment. They often have no effect, but they invade a microenvironment rapidly once the opportunity arises. In the mare this is often at covering, internal examination, AI, foaling, etc., especially at times of stress (Allen and Pycock, 1989). As such, they are potential causers of acute endometritis, especially in compromised or susceptible mares (Le Blanc *et al.*, 1991). There are three main opportunistic bacteria of concern.

Streptococcus zooepidemicus is implicated in 75% of acute endometritis cases, particularly during the initial stages. They are spherical bacteria, found normally in chain formation, often in the intestine and mucous membranes. Streptococcus is classified into two subgroups: alpha and beta. *S. zooepidemicus* is a beta streptococcus and, as such, causes the destruction of red blood cells and has a major role in initiating infection of the mare's cervix and uterus (Hughes and Loy, 1969). It may also promote the proliferation of other bacteria within the tract (Le Blanc *et al.*, 1991; Asbury and Lyle, 1993).

Haemolytic E. coli is a rod-shaped aerobic bacterium that is found either alone or in short chains. It is the second most common cause of uterine infection. It is naturally found in the intestine and is associated in particular with faecal contamination. It can cause not only acute endometritis but also severe systemic infection, which can prove fatal (Allen and Pycock, 1989; Asbury and Lyle, 1993).

Staphylococcus aureus is a less common cause of endometritis. It is a spherical or oval bacterium, normally evident in clusters and found associated with skin and mucous membranes. Under suitable conditions, such

as the disruption of the natural microflora, ill health or stress, the bacteria will invade the reproductive tract of the mare (Allen and Pycock, 1989; Asbury and Lyle, 1993).

19.3.2.5.3.1.2. Venereal-disease bacteria. Venereal-disease bacteria are those that are transferred solely via the venereal route, i.e. they are present within the semen and the reproductive tract of the mare and stallion and are capable of producing endometritis in both the normal and the susceptible mare. They may also be present in apparently asymptomatic animals – in particular, the stallion, which rarely shows symptoms. There are three main venereal-disease bacteria of concern.

Taylorella equigenitalis is an extremely contagious bacterium and is the causal agent of contagious equine metritis (CEM) (Tainturier, 1981). It was first isolated in Newmarket, UK, by Crowhurst (1977), where it spread rapidly and widely, due to the reluctance of infected-mare owners not to present their mares for service. The bacterium is initially a rod shape and becomes spherical with age. The stallion is seemingly not affected by the bacterium, but is the prime means by which it is spread from mare to mare. In the mare, the typical symptoms of acute endometritis are seen, characterized by uterine, cervical and vaginal inflammation, along with a copious grey discharge within 2–5 days of infection. In rarer instances, the mare may also not show any clinical symptoms but still be a carrier capable of infecting a stallion. At the other extreme, the infection may develop on to give chronic endometritis.

Klebsiella pneumoniae is an encapsulated rod-shaped bacterium, associated with acute and chronic endometritis. The ones of particular concern are capsular types 1, 2 and 5 (Pycock, 2000). The bacteria are endemic and widespread, but diagnosis is reasonably accurate via cervical–uterine swabbing. Unfortunately, the bacteria are relatively insensitive to antibiotics and antiseptic washing agents (Crouch *et al.*, 1972).

Pseudomonas aeruginosa, a slender rod bacterium with rounded ends and flagella, is

found widely within the environment. However, some strains of *P. aeruginosa* cause endometritis and may be isolated in stallion's semen or in swabs taken from the urethral fossa, but clinical symptoms are rarely evident. In the mare, *P. aeruginosa* causes a greenish-blue or yellowish-green exudate, which appears to be more prevalent in older mares. It is relatively resistant to antibiotics and antiseptics, so early diagnosis and cessation of natural cover is the best course of action (Hughes *et al.*, 1966; Hughes and Loy, 1975).

19.3.2.5.3.1.3. Diagnosis. Due to the highly contagious nature of venereal-disease endometritis, diagnosis and prevention are very important. Diagnosis of acute endometritis may be obvious due to exudates, or via identification of inflammation via scanning, rectal palpation or endoscopy. Once inflammation has been diagnosed, the causal agent needs to be identified by bacterial culturing. Bacterial culturing may also be carried out as a preventive measure.

Bacterial infections may be identified by swabbing of the reproductive tract. It is normal and recommended practice (Horse Race Betting Levy Board, 2001) that swabs are taken from the uterus, cervix, clitoris and urethra opening. Uterine swabbing should be carried out using a guarded swab to prevent contamination *en route* and through an open cervix during oestrus (Greenhof and Kenney, 1975; Fig. 19.8). The other swabs may be taken throughout the mare's oestrous cycle. The resultant swabs are plated out and incubated under varying conditions, normally aerobic and microphilic, to aid bacterial identification (Ricketts *et al.*, 1993). Fungal infections may also be identified in a similar manner. The use of swabbing is a widespread and often compulsory practice. Some breed societies have successfully used it to eradicate specific causes of infection in many areas worldwide. In particular, within the UK, the Horse Race Betting Levy Board publishes annual codes of practice, which are used worldwide and have resulted in a near eradication of *T. equigenitalis* (CEM) from the UK, as well as significantly reducing the incidence of venereal



Fig. 19.8. Many studs require all mares to be swabbed prior to service. The swabs are taken from clitoral sinuses and fossa, along with the urethral opening, endometrium and cervix.

diseases caused by *K. pneumoniae* and *P. aeruginosa*; it also produces guidelines on EHV and EVA (Horse Race Betting Levy Board, 2001). These codes of practice are reviewed annually and detail the number and type of swabs that need to be taken for different classes of mare. CEM is now a notifiable disease in the UK and codes of practice have been laid down for the exportation and importation of stock and, in cases of suspected CEM, abortion (Platt *et al.*, 1978; Crowhurst *et al.*, 1979; Rossdale *et al.*, 1979b).

Uterine aspirations and washings may also be collected, especially if purulent material and fluid are present. Culturing of the washings allows bacteria to be identified (Freeman and Johnston, 1987).

19.3.2.5.3.1.4. Acute endometritis. Acute endometritis is invariably a result of significant bacterial challenge by venereal or opportunistic bacteria. Acute infections develop rapidly, giving immediate symptoms of exudate or pus and irregular oestrous cycles. Internally, it causes deep haemorrhage and degeneration of luminal epithelial cells and, in severe cases, degeneration of the deeper stroma cells, leading to areas of missing endometrium. This may lead to hypertrophy and abscessed uterine glands (Rooney, 1970).

Acute endometritis is a major cause of infertility in the mare, providing a hostile environment for both sperm and embryo survival. Bacteria are introduced into the system at covering or at veterinary inspection. It is now accepted that some degree of acute endometritis is evident after all coverings, regardless of the extent of bacterial invasion. Normally, post-coital endometritis causes no long-term effect, the mare's system being able to adequately deal with the challenge presented, both via an immunological response and by uterine contractions used to eliminate infections and exudates. In most mares, the inflammatory response subsides within 72 h, and the uterus is able to receive the fertilized ovum. Occasionally, however, post-coital endometritis persists, becoming a major problem, with the potential of developing into a chronic infection (discussed later).

Treatment for general acute endometritis begins with an attempt to correct any physical abnormalities that may be predisposing the mare to infection, such as a Caslick vulvoplasty, a Pouret operation, removal of adhesions, etc. (Caslick, 1937; Pouret, 1982). The existing infection is then treated normally by local antibiotics, systemic antibiotics or uterine lavage (Threlfall, 1979). Local antibiotics are applied via infusion, using an indwelling catheter passed through the

cervix and placed in the uterus. The end of the catheter is looped into two ram's-horn shapes, which help keep the catheter in place and allow repeated infusions without the need to change and reintroduce the catheter (Figs 19.9 and 19.10). This reduces the risk of introducing more opportunistic bacteria via the technique itself into what is already a compromised system.

The catheter is most easily introduced through a relaxed cervix at oestrus. However, this is not always possible, as endometritis often leads to anoestrus and hence a tight cervix. Treatment with $\text{PGF}_{2\alpha}$ is therefore sometimes administered to bring the mare back into oestrus sooner, so reducing the delay until treatment can commence. $\text{PGF}_{2\alpha}$ may be administered as a series of injections to provide a series of short cycles and thus accelerate a series of treatments. Infusion times depend upon the severity of the infection and may vary from daily infusions for 3–5 days to 15 days. Such antibiotic treatment must be used with care, as some antibiotics may cause necrosis or erosion of the endometrium. Bacterial resistance may also be a problem, so identification of the causal bacterium and use of a specific target antibiotic is advocated. Excessive antibiotic use may allow fungal infections to develop, which will themselves require treatment (Asbury, 1987; Asbury and Lyle, 1993). Systemic antibiotics have been used, but evidence for their success is inconclusive. They have been advocated for use in conjunction with local antibiotics (Brown *et al.*, 1984).

Uterine lavage, using 1–2 l of saline, is increasingly popular. Lavage has been demonstrated not only to remove debris and exudate but also to encourage neutrophil release to the infection site. The washings

may also be used to identify causal agents. The extent and regularity of lavage again depends upon the severity of the condition (Asbury, 1990). Uterine infusion or lavage with chemical irritants or disinfectants, as a form of chemical curettage (discussed previously), has been advocated. However, results are variable and such treatments should be used with great care. Povidone-iodine has been used with some success (Asbury and Lyle, 1993), as has plasma infusion (Asbury, 1984).

Post-coital acute endometritis. Post-coital acute endometritis is the specific term given to acute uterine inflammation resulting from covering. As indicated previously, this is evident, to some extent, in all mares, but is especially evident in susceptible mares. The persistence of post-coital endometritis is encouraged by several factors, including general stress, a decline in the mare's general well-being and cervical, vaginal or vulval abnormalities. However, even in the absence of these predisposing factors, acute endometritis may still persist, and it is evident that some mares are inherently more susceptible than others. Initially, it was thought that the acute response in some mares was due to an immunological incompetence, rendering the mare unable to react to, and so eliminate, bacteria introduced at covering (Hughes and Loy, 1969). More recent research would indicate that there is no significant difference in immunoglobulin release or the functional ability of neutrophils in normal and susceptible mares *post coitum* (Allen and Pycock, 1989) and that an inflammatory response is evident even with the insemination of semen without bacterial contamination (Kotilainen *et al.*, 1994). It is

Fig. 19.9. An infusion catheter for the treatment of mares with antibiotic solution in cases of endometritis.



Fig. 19.10. An infusion catheter *in situ*, allowing repeated treatment of mares with antibiotics in cases of endometritis (photograph Elm Stud, Vikki Kingston).

now thought that acute post-coital endometritis is due to a reduction in the physical ability of susceptible mares to drain fluid from the tract. This is likely to involve defective myometrial contractility, but the cause of such a defect is as yet unclear (Troedsson and Lui, 1991; Le Blanc *et al.*, 1994).

Post-coital prophylactic measures are often employed in such mares to reduce the incidence of inflammation by assisting uterine exudate clearance. Those sperm required for fertilization reach the Fallopian tube within 2–4 h of ejaculation and the fertilized ovum does not arrive in the uterus until day 5 (Bader, 1982). In theory, therefore, uterine treatment is safe within these time limits. In practice, however, due to the rapid rise in progesterone after ovulation, it is best not to attempt treatment after 48 h post-ovulation. Treatment normally consists

of uterine lavage, using a saline plus antibiotic solution, which successfully removes uterine fluid and debris, enhancing neutrophil function and antibiotic efficiency. Lavage also stimulates uterine contractility and encourages the release of fresh neutrophils through irritation of the endometrium (Knutti *et al.*, 2000; Pycock, 2000). As such, it is often the treatment of choice for such mares. Oxytocin may also be used, both alone and in combination with lavage, again to encourage myometrial activity and hence fluid clearance (Allen, 1991; Le Blanc, 1994; Pycock and Newcombe, 1996). PGF_{2α} by virtue of its similar action on uterine myometrial contractility, has also been used successfully (Combs *et al.*, 1996). These systems may be supplemented by AI, with semen extended with antibiotic extenders. The addition of antibiotics significantly reduces any bacterial challenge and the use of AI reduces the total number of sperm introduced into the uterus and further reduces the inflammatory response (Davies Morel, 1999; Nikolakopoulos and Watson, 2000). For the same reason such mares should ideally only be covered once (Kenney and Ganjam, 1975; Kenney *et al.*, 1975a).

In the treatment of endometritis, topical treatments of vaginal or clitoral infections should be considered. This involves cleansing the whole area with a non-antiseptic soap for *K. pneumoniae* and *P. aeruginosa* or chlorhexidine for *T. equigenitalis*, followed by topical application of antibiotic creams. Unfortunately, *K. pneumoniae* and *P. aeruginosa* are particularly difficult to eliminate, in which case clitorectomy may be considered (Pycock, 2000). Clitorectomy – removal of the clitoris – is more widely practised in the USA than in the UK and Europe.

19.3.2.5.3.1.5. Chronic endometritis. Chronic endometritis may be more accurately divided into chronic infective endometritis and chronic non-infective degenerative endometritis. It has recently been suggested that chronic non-infective degenerative endometritis should now be termed endometrosis.

Chronic infective endometritis. Chronic infective endometritis can arise from an untreated or inappropriately treated acute uterine infection or due to a mare's inability to satisfactorily combat the initial infection. The condition is more often found in older multiparous mares, where the breakdown in uterine defence mechanisms results in an inability to respond to introduced infection and may also have allowed normal genital bacterial flora to contaminate the uterus. Such infection is often long-term, but is not so evident as a dramatic inflammatory response. It can be extremely damaging to the uterine tissue, causing degeneration and necrosis and resulting in permanent infertility. Treatment is as indicated for acute endometritis but with particular use of infusion and lavage. Large-volume infusion with a broad-spectrum antibiotic is advised, as often a wide range of bacteria are present (Pycock, 2000). Lavage using isotonic saline, followed by antibiotic and/or plasma infusion, is reported to be successful (Asbury and Lyle, 1993). At breeding, such mares should be treated in a similar manner to those susceptible to acute post-coital endometritis (Asbury and Lyle, 1993).

Endometrosis, or chronic non-infective degenerative endometritis. Endometrosis is caused by degeneration, rather than by infection of the endometrium, and may be classified as infiltrative or degenerative. Infiltrative endometrosis may be a result of changes within the uterus due to a busy breeding career, and is associated with a natural increase in leucocyte response to the normal bacterial challenge *post coitum* (Van Furth *et al.*, 1972; Ricketts, 1978). Degenerative endometrosis is a degeneration of the endometrial glands, rendering the uterus incapable of supporting a pregnancy. It is associated with EED and is often the result of repeated gestations, especially in mares with a history of uterine infections. Degeneration of the endometrial glands results in a failure to return to normal post-partum, leaving lymph-filled lesions. Treatment may be attempted by the stimulation of growth of new healthy endometrium using mechanical or chemical curettage, as

discussed previously. In particular, chemical curettage, using kerosene or povidone-iodine, followed by the infusion of antibiotics, has been used. However, such treatment is not very successful and runs the risk of further uterine damage. The prognosis for such mares is poor (Witherspoon, 1972; Bergman and Kenney, 1975; Kenney and Ganjam, 1975; Asbury and Lyle, 1993).

19.3.2.5.3.1.6. Acute metritis. Acute metritis is potentially the most serious uterine infection. It is associated with a massive contamination of the whole uterus as a result of trauma, often during parturition involving retained placental or fetal tissue, or due to bacterial infection introduced via air inspired post-partum or via hands used to aid parturition. Occasionally, it may be evident *post coitum*. Decomposition of retained tissue encourages rapid bacterial growth, along with toxin production. The inflammation of the entire uterus then favours the passage of toxins into the main circulation, resulting in toxæmia and, potentially, death.

Prevention is infinitely better than cure, and absolute hygiene at parturition, plus the complete expulsion of all placental and fetal tissue post-partum, is essential. Treatment must be immediate and normally involves large-volume lavage and possibly oxytocin to encourage uterine contraction and thus the flushing out of the uterine contents. Lavage should then continue until recovered fluids are relatively clear. Recovery is not possible until the source of the toxæmia is removed (Blanchard and Varner, 1993a; Threlfall, 1993). The prognosis is often poor and, even if the toxæmia is successfully resolved, long-term lameness from laminitis may result (Pycock, 2000; R. Eustace, Wales, 2001, personal communication).

19.3.2.5.3.1.7. Pyometra. Pyometra is characterized by fluid accumulation in a large, pendulous uterus. In time, the uterine walls may become leathery, tough and fibrous, due to continual infection. Such mares may appear healthy in themselves, but often do not show oestrous cycles, due to the inability of the uterus to produce PGF_{2α}. Pyometra

may be associated with a blockage of the uterus, fibrosis, adhesions, etc., resulting in a build-up of exudate within the uterus, with no normal drainage. It is often due to infection, but not necessarily so. Treatment normally involves drainage, followed by antibiotic infusion or lavage, but the prognosis for a breeding career is often poor. If infection is not evident and breeding is not required, such mares may not require treatment if they show no signs of discomfort. However, the presence of infection poses problems and, if left untreated, infective pyometra may develop into septicaemia (Ricketts, 1978; Hughes *et al.*, 1979).

19.3.2.5.3.2. Uterine fungal infections.

Mycotic or fungal infections of the endometrium are relatively uncommon in the mare. The most common is *Candida albicans*, a yeast. As with bacterial infections, they disrupt the ability of the uterus to support a developing embryo and, if present later in pregnancy, can cause abortion via placentitis and occasionally fetal infection. The majority of fungal abortions occur around 10 months of pregnancy (Platt, 1975; Acland, 1993). Treatment is via infusion of antimycotic agents, such as povidone-iodine or nystatin, but the success rate is low. Acidic agents, such as vinegar and acetic acid, have been used with some success (Pycock, 2000). If the mycotic growth cannot be arrested, the prognosis is hopeless.

19.3.2.5.4. CERVICAL INFECTIONS. Cervicitis, inflammation of the cervix, is usually associated with and is often the initial cause of endometritis. Such infection causes inflammation and possible pus accumulation (Sertich, 1993).

19.3.2.5.5. VAGINAL INFECTIONS. Vaginal infections are uncommon, the only one of note being dourine. Dourine is caused by a sexually transmitted protozoan, now eradicated from the UK, but still prevalent in many temperate countries. It causes vaginal and vulval infection and inflammation, along with discharge. If left untreated, it will develop systemically to form raised rings within the

mare's coat, along with depigmentation of the genitals, plus fever and death in 50–75% of cases (Brown, 1999).

19.3.2.5.6. VULVAL INFECTIONS. Equine coital exanthema, genital horse pox, evident as vesiculation and ulceration of the vulval lips or penis, is caused by EHV3. Infection causes pain at covering and can therefore affect reproductive performance. It is sexually transmitted and symptomless carriers are reported. There is no direct effect on reproductive performance, but, in order to prevent transfer, natural covering must cease. Treatment with antibacterial creams or powders prevents secondary infections and helps the natural healing process (Pascoe *et al.*, 1968, 1969; Gibbs *et al.*, 1972).

19.3.2.5.7. VIRAL INFECTIONS. The incidence of viral abortion is 1–5%, mainly occurring in late pregnancy. There are two viruses that have a major effect on reproductive performance in the mare: EVA and EHV.

EVA causes abortion in mares (Timoney and McCollum, 1987). The virus is spread via the respiratory route, via the venereal route and from the placenta of aborting mares (Acland, 1993). Stallions are the major routes of infection, seropositive stallions can be classified as shedders and non-shedders. It is the seropositive shedder stallions that are a risk to mares, and only seropositive mares should be put to them for covering. Alternatively, all mares should be vaccinated at least 3 weeks prior to breeding (Parlevliet and Samper, 2000). In the UK, EVA is a notifiable disease under certain circumstances and, as such, is now included in the Horse Race Betting and Levy Board *Codes of Practice*. It is as yet of minor concern in the UK.

There are four main strains of EHV: 1, 2, 3 and 4. EHV3 is the causal agent for equine coital exanthema, which has been discussed. EHV1 and EHV4 are of concern with regard to reproductive performance, as they can cause abortion. In particular, EHV1 is the causal agent for rhinopneumonitis abortion. Virus transfer is via the respiratory route, from birth fluids, soiled bedding, placental

tissue, etc. It may also be found in semen (Acland, 1993; Davies Morel, 1999). The virus causes placental separation resulting in fetal suffocation and abortion, with 96% occurring in the last 4 months of pregnancy. It can have a devastating effect, causing abortion storms in mares, plus neonatal losses. As with EVA, EHV1 is now included in the Horse Race Betting and Levy Board *Codes of Practice* and is as yet of minor concern in the UK.

19.3.2.6. Fetal congenital deformities

Many fetal developmental deformities have been reported (Liepold and Dennis, 1993). Many of these are not compatible with fetal

life and so cause abortion. Chromosomal defects also occur, leading to EED rather than abortion (Blue, 1981).

19.4. Conclusion

The causes of infertility, or the failure to produce an offspring, whether on a temporary or permanent basis, are numerous. Some are treatable but, for many, the prognosis for the individual as a breeding animal is poor. It is essential, therefore, that all potential breeding stock are submitted to a thorough examination prior to purchase, in order to ensure that they are capable of fulfilling their reproductive potential.

20

Artificial Insemination

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20.1. Introduction

Research specifically into artificial insemination (AI) in equids has been limited, as a number of breed societies will still not accept for registration progeny conceived in this way. The most noteworthy of these in Britain is the Thoroughbred Breeders Association, which in turn provides a large amount of

funding both directly and indirectly for equine research, especially related to reproduction. Most of the other breed societies within Britain and worldwide do now accept the progeny of AI, but many set strict regulations, such as a limit on the number of foals that can be registered per stallion per year. In Europe, Australia, China, South Africa and the USA, equine AI is now widespread in its

use. The historic opposition to AI has limited research work and hence the development of the technique. It therefore has some considerable way to go before it reaches the sophistication of cattle AI. More detailed accounts of equine AI may be found in Brinsko and Varner (1993), Davies Morel (1999) and Samper (2000).

20.2. Reasons for Using Artificial Insemination

There are a variety of reasons why equine AI is practised. Some of these are dependent upon and limited by the regulations set out by the countries and breed societies involved. The reasons for using AI include the following:

1. Removal of geographical restrictions.
2. Minimization of disease transfer, both venereal and systemic, by the removal of direct contact between the mare and stallion (Tischner, 1992). Semen may also be treated with extenders containing antibiotics to minimize the bacteria content and so reduce the number of potentially pathogenic organisms. Such semen is therefore useful for mares that have an increased susceptibility to uterine infection (Clement *et al.*, 1993).
3. Reduction in injury risk both to handlers and horses by the removal of direct contact between mare and stallion. The risk is further reduced if the stallion can be encouraged to mount a dummy mare.
4. Increasing the number of mares that can be inseminated per ejaculate.
5. Improvement of native stock through semen importation (Sukalic *et al.*, 1982; Ghei *et al.*, 1994).
6. Development of gene banks for future reintroduction of genetic material (Zafracas, 1994).
7. Breeding of difficult mares – those with physical abnormalities, especially caused by accidents, infection, poor perineal conformation, psychological problems, etc. However, care must be taken to ensure that such problems are not heritable.
8. Breeding from difficult stallions – those with physical problems, injury, infection, inadequate semen characteristics, psychological

problems, etc. (McDonnell *et al.*, 1991; Love, 1992). As with the mare care must be taken to ensure that such problems are not heritable.

9. Reduction in labour costs (Boyle, 1992).

10. Semen sexing (Johnson *et al.*, 1998; Seidel *et al.*, 1998; S. Revell, Wales, 2001, personal communication).

Several concerns over the use of AI have also been expressed, including the reduction of the genetic pool, overemphasis on 'fashionable' strains, the technical skill required and the infection risks if adequate screening is not practised (Davies Morel, 1999).

20.3. Semen Collection

Semen collection can be carried out using one of several methods. The easiest method is the collection of dismount samples. The drips of semen are collected from the stallion after withdrawal from the mare into a sterile jar. This method is unreliable, the quality of the sample is very variable and the majority of the sample is left within the mare. Samples often contain low sperm concentrations and are relatively high in pathogenic organisms. Semen can also be collected from the anterior vagina immediately after mating. However, the semen at collection has already come into contact with the acidic and hence spermicidal secretions found within the vagina. The sperm may also become contaminated by pathogenic organisms. Neither of these methods of collection allows assessment of the total volume of semen produced.

Condoms have been developed for use with horses. They can work very well, but do have a tendency to burst or become dislodged, resulting in the whole sample being lost (Perry, 1968; Boyle, 1992).

Finally, the best and now most commonly used method of semen collection is the artificial vagina (AV). The first AV was developed for use with horses in Russia at the beginning of the 20th century. Subsequent development of the AV was for use in cattle. Various models, including the Cambridge, Colorado, Missouri, Nishikawa and Hannover, are available, but they are all based on the same

principles (Davies Morel, 1999). They provide a warm, sterile lumen, surrounded by a water jacket, under some pressure, with a collecting vessel at the end, in an attempt to mimic the natural vagina (Figs 20.1 and 20.2).

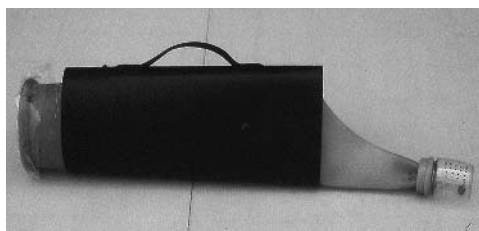
Most AVs consist of a solid outer casing with two rubber linings, an outer and an inner. The outer lining and the casing form a jacket, into which warm water and/or air is passed by means of a tap or valve. The water temperature inside the AV should be slightly above body temperature, at 44–48°C. The amount of water used must be adequate to ensure that the pressure within the lumen of the AV mimics, as closely as possible, the pressure against the insertion of the penis within the natural vagina. Some models (the Missouri) allow the lumen pressure to be increased by inflating the water jacket; as air is lighter than water, this minimizes the final weight of the AV. The inner lining of the AV

is often protected by an additional disposable inner liner, so ensuring sterility. The disposable liner, or in some cases the inner liner itself, is connected to the collecting vessel. Before use, this liner is lubricated with sterile obstetric lubricant to aid the stallion. It is most important that the collecting vessel, as well as the lumen of the AV, is about 44°C during collection, in order to prevent cold shock (Kayser *et al.*, 1992). Insulation and protection from ultraviolet light may be provided by enclosing the whole AV and collecting vessel within a protective jacket. The fully assembled AV prior to use is of the order of 50 cm in length and can weigh up to 10 kg (Fig. 20.3).

As discussed, sperm are susceptible to sunlight and temperature change. If the temperature of the AV or collecting vessel is too cold, sperm will suffer from cold shock and die (Kayser *et al.*, 1992). If the temperature of the AV is too hot, there is a similar detrimental effect on sperm. In addition, there is the risk of discouraging the stallion from using an AV, and also possibly from carrying out natural service. The stallion seems less sensitive to temperature than other farm livestock, but temperatures above 48°C must be avoided (Hillman *et al.*, 1980).



(a)



(b)



(c)



(d)

Fig. 20.1. Various artificial vaginas (AVs) for use in horses; all except the Nishikawa have disposable inner liners in place. (a) Cambridge AV; (b) Missouri AV; (c) Nishikawa AV; (d) Hannover AV.

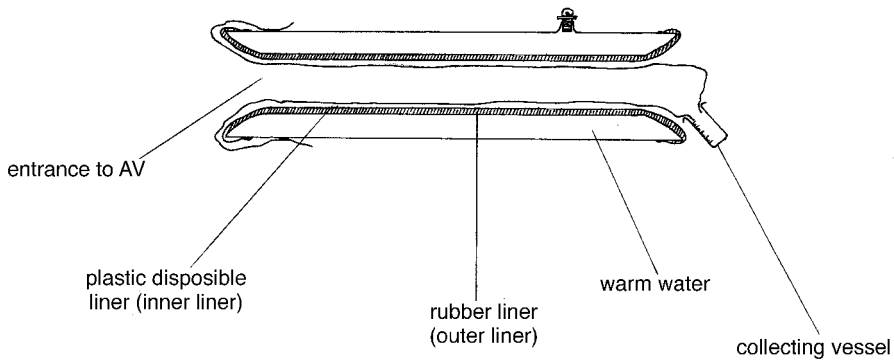


Fig. 20.2. A diagrammatic representation of an equine artificial vagina (AV).



Fig. 20.3. To protect the artificial vagina from ultraviolet light and to help maintain the required temperature, the whole thing can be enclosed within a protective jacket.

Stallions can be trained relatively easily to use an AV. Initial training uses an oestrous mare, prepared as for normal covering, to encourage mating and ejaculation into the AV. Use of an oestrous mare does, however, involve risks, not least her accidental covering. Many stallions are therefore trained to mount a dummy mare (Fig. 20.4).

Most stallions are quite happy to use such a dummy, especially if an oestrous mare is in the vicinity. Some stallions are not so keen, usually as a result of low libido, a possible consequence of coming into stallion work late in life, after time as a performance horse, or of incorrect AI management in the past.

These stallions may well require the extra stimulus of a jump mare. This mare may be a naturally occurring nymphomaniac that is in a continual state of oestrus due to hormonal imbalance or she may be treated with oestradiol-17 β to induce oestrus without ovulation. Not all nymphomaniac mares, however, are suitable as this condition can result in unpredictable behaviour.

A stallion is prepared for semen collection in the same manner as he would be for natural covering (see Chapter 13). A covering bridle should be used and semen may be collected in the normal covering yard or in a dedicated AI area. If a jump mare is to be used, she again is prepared as for natural service, including swabs in case accidental covering occurs. She must be of a quiet and calm disposition. This is especially important during the training of a young stallion (Fig. 20.5).

It is essential that all equipment is present and in satisfactory order and at the right temperature for both collection and subsequent handling before the stallion is brought in for collection. Everyone involved must know what is expected of him/her. Collection of semen always carries a risk, due to the unpredictable nature of stallions, especially when covering. As is regular with in-hand covering, all handlers are advised to wear hard hats, especially the semen collector. Up to three handlers will be needed if a jump mare is to be used, one to hold the stallion, one to collect the semen and one to hold the jump mare. All handlers should stand on the same side of the stallion. This

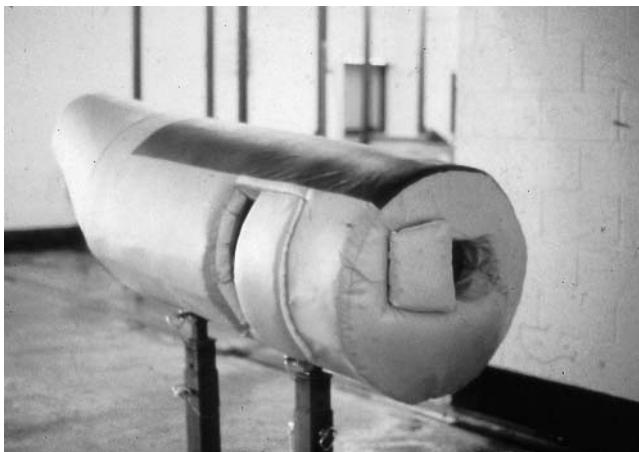


Fig. 20.4. Many stallions are trained to use a dummy mare. This eliminates the chance of accidental mating of a jump mare. (Thoroughbred Breeders Equine Fertility Unit, Newmarket, UK.)

ensures that, in the event of an accident, the stallion can be pulled away by his handler, and minimizes the chance of the semen collector getting kicked. The side used does not affect the sample collected, but, once a stallion has got used to semen being collected from one side, it is best to try and stick to that side in future.

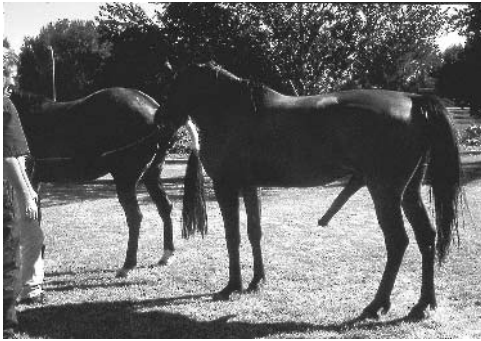
The stallion is allowed to mount and the collector diverts the penis, when erect, towards the AV. The stallion should be allowed to gain intromission and enter the AV of his own free will and not have the AV forced upon him. The AV can be stabilized by being held against the hindquarters of the mare, if present, or the back of the dummy (Fig. 20.5). The occurrence of ejaculation is noted, as in natural covering, by the flagging of the tail or by feeling for the contractions of the urethra and the passage of the semen along the ventral side of the penis.

After collection, the collecting vessel must be carefully removed from the AV and the semen evaluated as soon as possible. If it is not possible to carry out semen assessment immediately, it can be extended and stored at 4–5°C for up to 24 h without appreciable reduction in its viability, and a reasonably accurate evaluation of the semen can still be obtained (Malmgren *et al.*, 1994).

20.4. Semen Evaluation

Prior to evaluation, the gel fraction of semen has to be removed. This gel fraction is the later secretion and has a very low concentration of sperm, which are invariably dead. The gel fraction of semen can be removed by several methods: careful aspiration with a sterile syringe; filtration through a gauze or by an inline filtration system incorporated into the AV; or careful decanting. Filtration is the most popular method used today and also ensures that debris is removed (Davies Morel, 1999).

It is imperative that, at all times, including during handling and evaluation, semen is kept warm, at 38°C (slightly lower than the temperature actually at collection). Sperm are very susceptible to cold shock and, if any instruments, slides, microscope stages, etc. are not prewarmed, then results obtained will be misleading. Semen can be evaluated under several categories and not all evaluation involves all the assessments detailed below. It will vary according to what it is hoped to achieve by the assessment and the reasons for it being carried out. It is advisable for all the assessments to be carried out before a stallion enters an AI programme for the first time, and it is as well to repeat this at the beginning of every season. Appearance, motility, concentration and possibly morphology should be ideally assessed in each sample collected (Davies Morel, 1999).



(a)



(b)



(c)



(d)



(e)



(f)



(g)

Fig. 20.5. Events involved in semen collection using a jump mare: (a) introduction or teasing of the stallion and mare; (b) artificial vagina (AV) ready for use; (c) guiding the stallion's penis into the AV and the correct positioning of handlers on the same side of the mare and stallion; (d) slightly inclined positioning of the AV to mimic the natural position of the mare's vagina; (e) dismount of the stallion and slow removal of the AV, ensuring all semen is collected; (f) after dismount, the stallion should be turned away from the mare to reduce the chance of injury; (g) the collected sample (photographs Julie Baumber and Victor Medina).

When assessing the reproductive potential of a stallion, it is best to collect more than one sample for assessment. Two possible regimes are recommended for semen collection for evaluation: two ejaculates taken 1 h apart, followed 3 days later by a single ejaculate for evaluation; or two ejaculates collected 1 h apart, followed by daily collection of samples for evaluation for 6–7 days (Pickett and Voss, 1972; Swierstra *et al.*, 1975). However, these take time and are really not practical in many systems. In addition, any delays in testing are unpopular, as they delay the start of the breeding season. Therefore, in practice, most evaluation is carried out on a single ejaculation sample.

20.4.1. Gross evaluation

20.4.1.1. Appearance

Stallion semen is normally milky white in colour with a thickness equivalent to single cream (Kuklin, 1983; Fayrer-Hosken and Caudle, 1989). It should contain no evidence of bloodstaining, urine contamination or clots. If these are present, the sample should be discarded and the stallion examined (Varner and Schumacher, 1991; Samper, 1995a). The normal volume of semen produced by a stallion at each ejaculate varies considerably – 30–250 ml – but, on average, most stallions produce 100 ml and, of this, the gel fraction is normally 20–40 ml. However, considerable variation is evident, both between different stallions and between different collections throughout the year (Pickett *et al.*, 1988; Ricketts, 1993; Davies Morel, 1999). As discussed in Chapter 2, semen quality and quantity also vary within the breeding season.

20.4.1.2. pH

A standard pH meter can be used to assess the acidity/alkalinity of the semen sample. Acid conditions are known to be spermicidal; elevated pH may also be indicative of extraneous material or infection. A pH of 6.9–7.8 is acceptable, with levels of 7.3–7.7 being best (Pickett and Back, 1973; Fayrer-Hosken and Caudle, 1989; Oba *et al.*, 1993; Pickett, 1993a).

20.4.2. Microscopic evaluation

20.4.2.1. Semen extenders

Prior to microscopic evaluation, the sample is invariably extended, though an indication of motility is often assessed microscopically, using a raw sample. Dilution is primarily used to allow individual spermatozoon to be observed, as spermatozoa within raw semen tend to clump together, making more than a very rough estimate of motility impossible. Extension of the sample also prolongs the life of the spermatozoon, providing them with an additional source of energy and substrates for survival, and the maintenance of viability while evaluation takes place (Yates and Whitacre, 1988). Addition of the extender must be done immediately after collection, ideally within 2 min, to reduce the chance of obtaining erroneous results from a loss of spermatozoan viability caused by delay. There are numerous extenders available and used successfully during the evaluation process. In general, these are the same as those used in the preparation of semen for immediate AI and/or chilled storage. The most popular are those based upon either non-fat dried milk solids or skimmed milk (Table 20.1). The dilution rate required depends upon the concentration of sperm and also the test being performed.

Table 20.1. Examples of extenders used for semen evaluation.

Component	Quantity
Non-fat dried skimmed-milk (NFDSM) glucose extender (from Kenney <i>et al.</i> , 1975a)	
NFDSM	2.4 g
Glucose	4.0 g
Penicillin (crystalline)	150,000 units
Streptomycin (crystalline)	150,000 µg
Deionized water	Made up to 100 ml
Non-fortified skimmed milk extender (from Varner and Schumacher, 1991)	
Non-fortified skimmed milk	100 ml
Polymixin B sulphate	100,000 units
Heat milk to 92–95°C for 10 min in a double boiler, cool and add the polymixin B sulphate	

20.4.2.2. Motility

Sperm motility is graded on a scale of 0–5, 0 being very poor and 5 excellent. Motility must be assessed immediately in order to get an accurate measurement. It may be assessed visually or via a computerized motility analysis system. Undiluted semen can be examined visually under a light microscope. The wave movements of the sample are then graded. Alternatively, it can be assessed diluted, when the motility characteristics of individual sperm can be graded, although dilution itself may affect motility. Assessment therefore tends to be subjective and dependent on the examiner's previous experience (Jasko *et al.*, 1988). Table 20.2 gives a rough guide to the grades (0–5) often used.

Due to the subjectivity of microscopic assessment, other methods of assessing motility have been investigated, including the time-lapse dark-field photographic method (Elliott *et al.*, 1973; van Huffel *et al.*, 1985), cinematographic techniques (Plewinska-Wierzbowski and Bielanski, 1970) and computer analysis (Jasko, 1992; Burns and Reasner, 1995). The correlation between the results obtained by visual and computer analysis is reported to be very good, at 0.92 (Bataille *et al.*, 1990; Malmgren, 1997). Computer analyses have now been developed to carry out a full evaluation, not just of motility (Jasko *et al.*, 1990b). The cost of the equipment means that such analyses are largely restricted to research use, though they do provide an automated, rapid, objective result.

Regardless of assessment method used, classification of type of movement, as indicated in Table 20.2, is important. Progressive movement is required; oscillatory movement (movement on the spot) or movement in very tight circles is classified as abnormal. If individual sperm are not assessed, then the wave patterns of undiluted sperm can give only an indication of individual sperm movement. Some evaluators classify sperm motility as a ratio of those showing progressive movement : oscillatory movement.

Semen containing at least 40% actively progressively motile spermatozoa, which is grade 2–3, can be considered adequate for AI (Colenbrander *et al.*, 1992; Ricketts, 1993; Davies Morel, 1999). The correlation between motility and fertility is, however, variable and low (0.7) (Pickett *et al.*, 1975b; Samper *et al.*, 1991; Jasko *et al.*, 1992b; Heitland *et al.*, 1996). Though it gives the best correlation to fertility compared with all other parameters, it cannot be relied upon to give an absolute indication.

20.4.2.3. Longevity

Assessment of motility over time may be used as an indication of viability. However, it must be remembered that longevity in a test-tube does not necessarily equate to longevity within the mare's uterus. Attempts have been made to mimic longevity in the mare's tract (Kenney *et al.*, 1975a). Longevity assessment involves the evaluation of initial motility and then at various intervals after storage

Table 20.2. Criteria for grading sperm motility.

Grade	Description
0	Immotile
1	Stationary or weak rotatory movements
2	Backward and forward movement or rotatory movement, but fewer than 50% of cells are progressively motile and there are no waves or currents
3	Progressive, rapid movement of sperm with slow currents, indicating that about 50–80% of the sperm are progressively motile
4	Vigorous, progressive movement with rapid waves, indicating that about 90% of the sperm are progressively motile
5	Very vigorous forward motion with strong, rapid currents, indicating that up to 100% of the sperm are progressively motile

at 5°C, 22°C or 37–38°C. For example, if motility is not less than 45% after 3 h or 10% after 8 h at 22°C, then the semen may be classified as good enough for AI (Kenney *et al.*, 1975a; Amann, 1988; Amann *et al.*, 1988; Samper, 1995b).

20.4.2.4. Concentration

Sperm concentration within a semen sample is a major determinant of its value and was traditionally assessed using a haemocytometer. Manual or automated spectrophotometers are a more recent development and may now form part of a complete computer sperm-analysis system (Davies Morel, 1999). A haemocytometer consists of a counting slide with a cover slip, under which a set volume of diluted semen can be trapped and viewed. The number of sperm within the sample on the haemocytometer is counted by means of the counting grid. From this figure and the dilution rate, the concentration of sperm can be calculated. This method gives a very accurate assessment, but is time-consuming and cannot be easily done in the field. The spectrophotometer, on the other hand, can be used anywhere, but the results obtained can be variable and only as good as the initial calibration using a haemocytometer. The semen is diluted to approximately 1:30; the diluent used must be optically clear – for example, 10% formalin plus 0.9% saline. The amount of light passing through the sample is then read by the spectrophotometer, from which the concentration of sperm is calculated (Pickett and Back, 1973; Bielanski, 1975; Hurtgen, 1987; Davies Morel, 1999).

The normal range for sperm concentration is $30\text{--}600 \times 10^6$ sperm ml^{-1} of undiluted semen but concentrations do vary significantly (Pickett and Voss, 1972). A total number of 100×10^6 viable sperm per inseminate is required for acceptable fertilization rates (Kenny *et al.*, 1971). In practice, samples with a concentration of $100\text{--}200 \times 10^6$ ml^{-1} are considered acceptable for AI (Pickett *et al.*, 1988; Ricketts, 1993; Davies Morel, 1999).

20.4.2.5. Morphology

Morphological examination is a common method of trying to assess a stallion's fertility. Sperm in a diluted semen sample are examined microscopically. Individual sperm are examined and the percentage of abnormal sperm in the sample noted. Viewing of individual sperm can be enhanced using a variety of stains, which allow various spermatozoan components or abnormalities to be identified (Reifenrath, 1994; Samper, 1995a), e.g. nigrosin-eosin stain, which stains the heads of dead sperm violet/purple (Dott and Foster, 1972). Staining of sperm to assess the integrity of specific areas may also be carried out, e.g. acrosome stains (Oetjen, 1988), immunofluorescence tests (Zhang *et al.*, 1990a) and labelled monoclonal antibodies (Blach *et al.*, 1988). The principles of these methods have been exploited in automated spermatozoa morphometry analysis systems (Gravance *et al.*, 1996). However, such systems are specialist and expensive and, as such, do not often form part of the routine evaluation process. Figure 20.6 illustrates some examples of the abnormalities that may be seen.

Abnormalities can be classified as primary (failure of spermatogenesis, failure of sperm maturation), secondary (sperm damage occurring at ejaculation) and tertiary (inappropriate handling after ejaculation). Failure of spermatogenesis is usually characterized by sperm with two heads, two tails, no midpiece, no tail, rudimentary tails or excessively coiled tails. Such abnormalities may be indicative of a long-term or even permanent problem. Maturation failure is characterized by the presence of cytoplasmic droplets on the midpiece of the sperm. As maturation proceeds, these cytoplasmic droplets progressively move down the tail and disappear. If they are present in ejaculated sperm, it is an indication that the sperm are immature. This may well be only temporary, a period of rest rectifying the problem. Damage occurring at ejaculation is normally characterized by tail abnormalities, bends, coils, kinks or swellings, detached heads and tails and protoplasmic droplets. Finally, post-ejaculatory damage is manifest as a loss of acrosome, fraying/thickness of

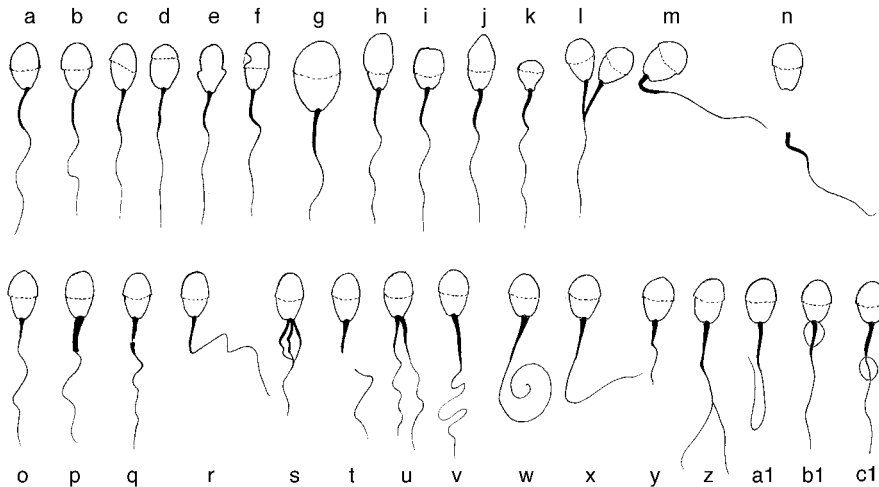


Fig. 20.6. Some examples of the more common abnormalities that may be seen when examining sperm for morphology. Top row from left to right: (a) a normal spermatozoon is followed by the following abnormalities in order. *Acrosome defects*: (b) swollen, (c) partially lifted, (d) small, (e) lifted, (f) part missing; *head defects*: (g) big head, (h) elongated, (i) flattened, (j) lanciolated, (k) microhead, (l) double head; *neck defects*: (m) bent, (n) broken. Second row: *mid-piece defects*: (o) short, (p) fat, (q) split/constricted, (r) bent annulus, (s) fibrous, (t) broken, (u) double; *tail defects*: (v) convoluted, (w) corkscrew, (x) bent, (y) small, (z) double, (a1) shoehorn; *droplets*: (b1) proximal, (c1) distal.

the midpiece and the bursting of sperm heads (Hurtgen, 1987; Varner *et al.*, 1987; Yates and Whitacre, 1988).

The correlation between morphology and fertility is relatively low (0.25–0.5) (Van Duijn and Hendrikse, 1968; Hurtgen, 1987). Work has been done in an attempt to correlate specific abnormalities to fertility, but again correlations are poor. Morphology, therefore, is not a very good indicator of the fertilizing potential, though, as with motility, in the absence of anything else, it is used.

Semen containing 65% or more morphologically normal sperm is considered appropriate for AI (Fayrer-Hosken and Caudle, 1989; Pickett, 1993a; Davies Morel, 1999).

20.4.2.6. Live : dead ratio

Motility gives an indication of the percentage of live sperm within a sample. However, differential staining of sperm, which are then examined microscopically, gives a more accurate result. Staining with nigrosin–eosin in equal volume to that of semen allows the differential staining of dead and live sperm. Dead sperm, as they

are permeable to the stain, appear as violet/purple (Dott and Foster, 1972, 1975). Other stains have been used equally successfully, including ethidium bromide and acridine orange fluorescence or H25 fluorescence (Hermetet *et al.*, 1993).

The ratio, or percentage, of dead to live sperm is assessed in a number of samples from the collection. A live : dead ratio of 6 : 4 (60% live sperm) is acceptable for AI. However, a sample with a lower ratio may be considered to be usable, if it has a high total sperm count, as it can be diluted less and this still ensures that the minimum number of live sperm for fertilization are inseminated (Ricketts, 1993; Davies Morel, 1999).

20.4.2.7. Cytology

Blood cells, leucocytes and erythrocytes can be identified in a semen sample, using haematoxylin and eosin or Wright's stain, and viewed under a haemocytometer (Roberts, 1971b; Swerczek, 1975). A count of leucocytes greater than 1500 ml^{-1} is indicative of a problem, possibly due to an infection, especially if the pH of the semen is also high. Such a

sample would not be appropriate for use in AI (Rossdale and Ricketts, 1980; Ricketts, 1993; Davies Morel, 1999). Erythrocyte concentrations above 500 ml^{-1} are indicative of a problem, such as haemorrhage, injury, etc., and would also make a sample inappropriate for use in AI (Rossdale and Ricketts, 1980; Ricketts, 1993; Davies Morel, 1999).

20.4.2.8. Bacteriology

Semen samples will potentially contain bacteria, both pathogenic and non-pathogenic (Madsen and Christensen, 1995). Isolation and identification of pathogenic bacteria, in particular, is required in order to prevent passage of infection at insemination. Bacteria can be identified by direct plating of semen samples on to agar plates or by plating of swabs taken from semen or the genitalia. Incubation under various conditions allows differentiation of bacteria (Madsen and Christensen, 1995). Long-term or acute infection may also be evident as high leucocyte counts in a semen sample or the evidence of pus. Identification of bacteria such as *Pseudomonas aeruginosa*, *Taylorella equigenitalis* and *Klebsiella pneumoniae* is likely to preclude a semen sample from use (Fraser, 1986; Scherbarth *et al.*, 1994; Davies Morel, 1999).

A summary of the acceptable semen parameters is given in Table 20.3.

All the tests indicated above are based upon gross or microscopic evaluation. The future of accurate semen evaluation may lie in functional tests. Many have been investigated, but none is reliable enough as yet to be used commercially (Wilhelm *et al.*, 1996).

20.5. Semen Storage and Use

Once the semen has been evaluated, it can be considered for insemination. Semen can be used in one of four ways: first, used immediately undiluted to inseminate a single or possibly two mares, depending on the volume collected; second, diluted and used immediately for insemination into several mares; third, diluted and refrigerated for use over the next 72 h; or fourth, diluted and frozen for use at a later date. The method used depends on the stud system and the location of the mare(s) to be inseminated.

20.5.1. Raw semen

The best results are obtained using undiluted semen inseminated immediately. However, such use of semen defeats two of the main objectives of AI – increasing the number of mares that can be fertilized with one ejaculate and transportation – though it may be of use as a veterinary or management aid.

20.5.2. Chilled semen

If semen is to be extended prior to use, it should be done so immediately (Ginther, 1992). A vast array of diluents have been developed, based normally upon milk, gelatin or egg yolk plus antibiotics (Squires *et al.*, 1981b; Varner, 1991; Jasko *et al.*, 1992a; Ijaz and Ducharme, 1995; Lawson and Davies Morel, 1996). Table 20.4 gives five of the more popular ones.

Table 20.3. The acceptable range for a normal stallion's semen parameters.

Parameter	Acceptable range
Volume of sperm produced	30–250 ml
Sperm concentration	$30\text{--}600 \times 10^6 \text{ ml}^{-1}$
Morphology	Minimum 40–50% physiologically normal
Live : dead ratio	6.0 : 4.0
Motility	Minimum 40% progressively motile sperm
Longevity at room temperature	45% alive after 3 h 10% alive after 8 h
pH	6.9–7.8
White blood cells	$< 1500 \text{ m}^{-1}$
Red blood cells	$< 500 \text{ ml}^{-1}$

Table 20.4. Examples of semen extenders used for chilled semen.

Component	Quantity
E-Z Mixin (from Province <i>et al.</i>, 1984, 1985)	
Non-fat dry milk	2.4 g
Glucose monohydrate	4.9 g
Sodium bicarbonate (7.5% sol.)	2.0 ml
Polymixin B sulphate (50 mg ml ⁻¹)	2.0 ml
Distilled water	92.0 ml
Osmolarity (mOsmol kg ⁻¹)	375.00 ± 2
pH	6.99 ± 0.02
Mix the liquids first and then add the powders	
Non-fat dried skimmed-milk (NFDSM) glucose extender II or Kenney extender (from Kenney <i>et al.</i>, 1975a)	
NFDSM	2.4 g
Glucose	4.9 g
Gentamicin sulphate (reagent grade)	100.0 mg
8.4% NaHCO ₃	2.0 ml
Deionized water	92.0 ml
Mix the liquids first before adding the NFDSM, to avoid the antibiotic curdling the milk	
Skimmed milk gel extender (from Voss and Pickett, 1976; Householder <i>et al.</i>, 1981; Pickett, 1993b)	
Skimmed milk	100.0 ml
Gelatin	1.3 g
Add the gelatin to the skimmed milk and agitate for 1 min	
Heat mixture in boiler for 10 min at 92°C, swirling mixture periodically	
Cream gel extender (from Voss and Pickett, 1976; Lawson, 1996)	
Gelatin	1.3 g
Distilled water	10.0 ml
Half-and-half cream	90.0 ml
Penicillin	100,000 iu
Streptomycin	100,000 iu
Polymixin B sulphate	20,000 iu
Osmolarity (mOsmol kg ⁻¹)	280.00
pH	6.52
Add gelatin to distilled water and autoclave for 20 min. Heat cream in boiler at 92–95°C for 10 min and add cream to gelatin solution, after removing scum from heated cream, to make a total of 100 ml	
CGH-27 extender (from Lawson, 1996)	
Glucose	4.5 g
Glycine	0.7 g
Casein	0.5 g
Egg yolk	7.0 ml
Gelatin	0.1 g
Sodium phosphate	0.05 g
Potassium chloride	0.025 g
Sodium potassium tartrate	0.25 g
Distilled water	100.0 ml
Osmolarity (mOsmol kg ⁻¹)	305.00
pH	6.72

All these extenders give acceptable fertilization rates for chilled semen. Extended semen can be stored for up to 2–3 days if extended in a ratio 2:1, semen:extender and cooled slowly to 4–8°C over 4 h and kept at this temperature until use (Hughes and Loy, 1970; Allen *et al.*, 1976a; Francel *et al.*, 1987; Malmgren *et al.*, 1994). Such treatment allows limited semen transportation and storage in a refrigerator or Equitainer (Fig. 20.7), as long as the temperature is kept at 4°C. The Equitainer is so arranged to ensure a gradual drop in semen temperature of $-0.3^{\circ}\text{C min}^{-1}$ down to 4–5°C. It will then keep the semen at this temperature for up to 24–48 h (Douglas-Hamilton *et al.*, 1984).

20.5.3. Frozen semen

Prolonged storage can only be achieved by freezing, but the techniques of freezing horse semen are nowhere near as refined as those for cattle (Amann and Pickett, 1987; Pickett and Amann, 1993). Numerous extenders have been tried, based upon those indicated in Table 20.5, most are similar to those used

for chilled insemination, but with the addition of a cryoprotectant, such as glycerol. The major problem is identifying a suitable cryoprotectant that can be used to prolong the formation of ice crystals either within the sperm head, so reducing physical damage, or within the surrounding solution, reducing sperm desiccation. Glycerol is used as such an agent in cattle, but appears to be toxic to equine sperm (Klug *et al.*, 1975; Nishikawa, 1975; Pace and Sullivan, 1975; Tischner, 1979; Fahy, 1986; Heitland *et al.*, 1996). However, in the absence of any other successful agent, low concentrations of glycerol continue to be used (Piao and Wang, 1988). Detergents and a combination of sugars have also been used as cryoprotectants (Naumenkov and Romankova, 1981, 1983; Cochran *et al.*, 1984; Piao and Wang, 1988). Examples of such extenders are given in Table 20.5. In addition to variation in results with extenders, there is great variation between and within stallions, with pregnancy rates of 10–70% being reported (Muller, 1987; Thomassen, 1991; Wockener and Colenbrander, 1993).

20.6. Semen Dilution

The extent of semen dilution depends on the initial concentration of the sample and the motility of the sperm (Magistrini *et al.*, 1987). Insemination of diluted semen containing 100×10^6 progressively motile sperm (PMS) per insemination gives good results, but normally 500×10^6 sperm per insemination is recommended in order to allow a margin of error (Pickett and Back, 1973; Cooper and Wert, 1975; Pace and Sullivan, 1975; Householder *et al.*, 1981; Piao and Wang, 1988; Vidament *et al.*, 1997). The insemination volume is calculated using the following formula:

Insemination volume (ml) =

$$\frac{\text{Number of PMS required}}{\text{Number of PMS ml}^{-1}}$$

$$\text{PMS ml}^{-1} = \% \text{ PMS} \times \text{number of sperm ml}^{-1}$$

$$\text{PMS required} = 100\text{--}500 \times 10^6$$

The insemination of 800×10^6 sperm is advised when using frozen semen, in order to compensate for loss occurring during the freezing process (Samper, 1995b).



Fig. 20.7. The component parts of the Equitainer. The ballast bags can be seen in the foreground; behind them are the two coolant cans or blocks and to their left the black foam insulating jacket (which acts as a thermal impedance pad and foam-rubber protective padding) into which the plastic semen container (isothermalizer) – far left – is placed. (From Hamilton-Thorn Research, Danvers, Massachusetts.)

Table 20.5. Extenders used in the cryopreservation of equine semen.

Component	Quantity
Skimmed-milk cryopreservation extender (from Samper, 1995b)	
Skimmed milk	2.4 g
Egg yolk	8.0 ml
Sucrose	9.3 g
Glycerol	3.5%
Distilled water	100.0 ml
Cryopreservation extenders incorporating detergent (Equex STM, Nova Chemicals Sales, Scituate, Massachusetts) used for the cryopreservation of stallion semen (from Cochran <i>et al.</i> , 1984)	
Lactose solution (11% weight : volume)	50.0 ml
Glucose EDTA solution (EDTA glucose extender II primary extender)	25.0 ml
Egg yolk	20.0 ml
Glycerol	5.0 ml
Equex STM	0.5 ml
Lactose-, mannitol- and glucose-based cryopreservation extender (from Naumenkov and Romankova, 1981, 1983)	
Lactose	6.6 g
Mannitol	2.1 g
Glucose	0.7 g
Disodium EDTA	0.15 g
Sodium citrate dihydrate	0.16 g
Sodium bicarbonate	0.015 g
Deionized water	100.0 ml
Egg yolk	2.5 g
Glycerol	3.5 ml

EDTA, ethylenediamine tetra-acetic acid.

20.7. Insemination Volume

The volume of inseminate varies from 10 to 30 ml for fresh semen, 10 to 60 ml for chilled and 0.5 to 5 ml for frozen (British Equine Veterinary Association, 1997). It has been suggested that volumes in excess of 100 ml or less than 0.5 ml are detrimental to conception rates (Rowley *et al.*, 1990; Jasko *et al.*, 1992b).

20.8. Insemination Technique

Mares are inseminated non-surgically. When using fresh or chilled semen, this should occur, as with natural service, on either day 2 or day 4 of oestrus (Hyland and Bristol, 1979; Voss *et al.*, 1979; Katila *et al.*, 1996; Watson and Nikolakopoulos, 1996). Frozen semen requires better synchrony with ovulation, ideally to within 6 h (Heiskanen *et al.*, 1994a, b; J. Newcombe, Wales, 2001, personal communication; S. Revell, Wales, 2001, personal

communication). Semen, both diluted and undiluted, is deposited into the uterus by means of a plastic sterile pipette, with syringe attached, or by an insemination gun, guided in through the cervix to the uterus, using the index finger. Alternatively, the pipette can be guided in through the cervix as per rectal palpation, the cervix being felt through the rectum wall (Fig. 20.8).

The syringe is loaded with semen held between two air bubbles and then attached to the end of the pipette. Once through the cervix, the insemination pipette is pushed into the uterus about 2 cm. When it is in place, the semen is slowly expelled by depressing the plunger (Figs 20.9 and 20.10; Davies Morel, 1999).

20.9. Conclusion

In many parts of the world, equine AI is widespread in its use. The UK, though a

Fig. 20.8. Artificial insemination in the mare.

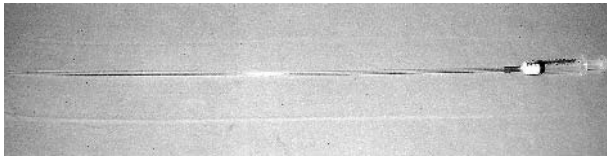


Fig. 20.9. In readiness for insemination, the filled syringe is attached to the end of the insemination pipette (photograph Julie Baumber and Victor Medina).



Fig. 20.10. Once the hand is lubricated, it should be introduced slowly into the vagina along with the insemination pipette. Once in place, the plunger of the syringe should be slowly depressed to expel all the semen into the uterus (photograph Julie Baumber and Victor Medina).

leader in many aspects of the equine industry, lags behind, largely due to the failure of the Thoroughbred industry to recognize and hence register progeny conceived by AI. Until it can be persuaded that AI is an acceptable means of breeding the horse, the

application of AI will be restricted to use in other breed societies. Despite this, it is evident that equine AI is here to stay and will continue to expand, opening up with it exciting opportunities in the selection and breeding of the equine species.

21

Embryo Transfer in the Mare

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21.1. Introduction

The first reported successful equine embryo transfer (ET) was carried out surgically in Japan in 1974 (Oguri and Tsutsumi, 1974). Research since then, including the development of non-surgical techniques has considerably improved the early low success rates (McKinnon and Squires, 1988). Nevertheless, the commercial application of ET in horses has a long way to go before it reaches the sophistication and success of its application in cattle and sheep. One of the major constraints on the development of ET in horses is the continued reluctance of breed societies to register foals conceived in this manner.

21.2. Reasons for Using Embryo Transfer

ET may be used for a number of reasons, including the following:

1. To obtain foals from mares that are unable to carry a foal to term or to go through the process of parturition.
2. To obtain foals from older mares without risk.
3. To provide a genetically promising foal with the best maternal environment, both intra- and extra-uterine (maximum milk production).
4. To allow performance mares to breed without interrupting their performance career.

5. To provide embryos for freezing and so provide genetic diversity in the future.
6. To aid in the breeding of exotic equids.
7. To increase the number of foals per mare per lifetime.
8. For biotechnology.
9. To allow cloning, embryo sexing, etc. (Wagoner, 1982; Ginther, 1992; Warren Evans, 1992).

Several concerns have been expressed with regard to the technique, including fear over the economic effect on certain sections of the industry, the possibility of inbreeding and reduction in the genetic pool and the cost of the procedure (East *et al.*, 1999b, c).

21.3. Donor and Recipient Mares

The main principle behind ET is the transfer of elite embryos from a genetically superior donor mare mated to a genetically superior stallion into a normally, but not necessarily, genetically inferior recipient mare that is reproductively competent. This technique makes use of the fact that the genetic make-up of the mare carrying the foal has no effect whatsoever on the characteristics of that foal. The foal's genetic make-up and therefore its characteristics are determined by the mare that produced the ovum and the stallion whose sperm was used to fertilize it. The technique also makes use of the fact that, for the first 20 days of its life, the equine embryo is free-living within the uterus and has not yet formed an attachment. Therefore, moving it to another mare's uterus can be carried out with reasonable ease.

In order for ET to be successful, the stage of the uterus into which the embryo is transferred must be synchronized with that of the uterus from which it was collected. This will ensure that the uterine secretions and development match the requirements of the embryo. To achieve this, the oestrous cycles of the donor and recipient mares must be synchronized. This is normally achieved by using exogenous hormone therapy. Further details on the means of synchronizing and timing oestrus and ovulation in the mare are given in Chapter 12. Initial evidence sug-

gested that the donor and recipient mares should ovulate within 24 h of each other, but more recent research has reported that success can be achieved if donors ovulate between 3 days before and 2 days after the recipient. The best results, however, are achieved if the recipient mare ovulates 24 h after the donor mare, thus allowing compensation for any developmental retardation that may occur to embryos undergoing the stress of transfer (McKinnon *et al.*, 1988c; Squires, 1993d).

21.4. Hormonal Treatment of Donor and Recipient

For ET, both the donor and recipient mares are similarly synchronized, normally using prostaglandin $F_{2\alpha}$ ($PGF_{2\alpha}$), commercially available as Equimate (Palmer and Jousset, 1975). Human chorionic gonadotrophin (hCG) may also be used in combination with $PGF_{2\alpha}$ to further synchronize and hasten ovulation (Allen *et al.*, 1976b; Voss, 1993).

21.4.1. The donor mare

In an ideal transfer system, a large number of embryos are collected on one occasion from a donor. She is induced to produce many more embryos than she would during her natural oestrous cycle – that is, she is superovulated. In cattle and sheep, superovulation is quite successful, though results do vary. Pregnant mare serum gonadotrophin, used as a superovulation agent in cattle and sheep, has no effect in mares. Some success has been reported in mares using equine pituitary extract, known commercially as Pitropin (Douglas *et al.*, 1974; Lapin and Ginther, 1977; Douglas, 1979). Daily injection over 7 days increases ovulation rates, but not consistently either between different oestrous cycles in the same mare or between mares. Experiments are at present being conducted into the use of gonadotrophin-releasing hormone (GnRH) and appear reasonably promising, though again this involves a series of injections

(Harrison *et al.*, 1991). Further along these lines, some success has been achieved using human menopausal gonadotrophins (Koene *et al.*, 1990) and porcine follicle-stimulating hormone (pFSH) (Fortune and Kimmich, 1993). However, the failure to find a reliable superovulating agent remains a major stumbling block in the development and commercial use of equine ET.

A general routine that may be used to synchronize and attempt to superovulate a donor mare is given in Table 21.1. In practice, Pitropin is not used; however, the regime indicated in Table 21.1 is still used, but with the omission of Pitropin (Bristol, 1981). As indicated, hCG may be used. hCG has properties similar to those of luteinizing hormone (LH) and can therefore be used to induce ovulation. However, mares treated repeatedly with hCG develop antibodies, which are reported by some to cause refractoriness (Wilson *et al.*, 1990), though this effect is disputed by others (J. Newcombe, Wales, 2001, personal communication). GnRH is therefore becoming an increasingly popular alternative to hCG, as it encourages the release of endogenous LH and FSH and does not cause an antibody response (Mumford *et al.*, 1995).

Table 21.1. A general hormone routine that can be used to time and attempt to superovulate a donor mare for embryo transfer. In practice, no attempt is made to superovulate; therefore, Pitropin is not used.

Time	Drug to be administered/event
Day 0	PGF _{2α} (Equimate)
Day 6	hCG
Day 10	Oestrus and ovulation in some mares
Day 14	Equimate and Pitropin
Day 15	Pitropin
Day 16	Pitropin
Day 17	Pitropin
Day 18	Pitropin, oestrus may start
Day 19	Pitropin, oestrus
Day 20	Pitropin, oestrus
Day 21	hCG, oestrus
Day 22	Ovulation may occur – covering/Al
Day 24	Ovulation may occur – covering/Al
Day 30	Embryo collection
Day 31	Embryo collection

PGF_{2α}, prostaglandin F_{2α}; hCG, human chorionic gonadotrophin; Al, artificial insemination.

21.4.2. The recipient mare

The ideal recipient mares are 5–10 years of age, with a proved breeding record and no history of uterine infection. They need be of no particular genetic merit, as they will in no way affect the genetic make-up of the embryos transferred to them. Ideally, the recipient mare should be larger than the donor, providing a larger uterus and so maximizing fetal advantage (East *et al.*, 1999a,c).

Recipients may be treated in a very similar manner to donors, except no attempt is made to superovulate them and they are, of course, not mated. An example of an exogenous hormone treatment regime used in recipients is given in Table 21.2.

They should be teased, rectally palpated or scanned to ensure that they have reacted to the synchronization programme and ovulated. Ovariectomized mares have been used experimentally as recipients. These mares alleviate the need for synchronization, teasing and veterinary inspection, but, because of the lack of ovarian progesterone, they require artificial progesterone supplementation for the first 120 days of pregnancy (Hinrichs and Kenney, 1988; Squires *et al.*, 1989).

21.5. Embryo Recovery

Once ovulation in the donor is confirmed, she is either covered naturally or artificially inseminated (AI). The age of embryos recovered varies with the chosen method of recovery. Equine embryos in general are collected at between 4 (morula) and 8 days (blastocyst) of age (see Figs 5.2 and 5.4). They can be recovered at a relatively late stage compared with other farm livestock, as equine conceptuses do not expand into elongated trophoblasts but remain spherical and free-living for a prolonged period.

21.5.1. Surgical recovery

The initial method of recovery during early work was surgical. This technique is still used by some and results in similar, or possibly

Table 21.2. An example of a hormone regime used to time ovulation in a recipient mare.

Time	Drug to be administered/event
Day 0	PGF _{2α} (Equimate)
Day 6	hCG
Day 10	Oestrus and ovulation in some mares
Day 14	Equimate
Day 18	Oestrus may start
Day 19	Oestrus
Day 20	Oestrus
Day 21	hCG, oestrus
Day 22	Ovulation may occur
Day 24	Ovulation may occur
Day 30	Embryo transfer
Day 31	Embryo transfer

PGF_{2α}, prostaglandin F_{2α}; hCG, human chorionic gonadotrophin.

lower, recovery rates compared with the more recently developed non-surgical techniques. Surgery is carried out under general anaesthetic, with the mare presented in a dorsal recumbency with her ventral side (abdomen) uppermost. The uterus is exteriorized through a ventral midline incision in the area between the mammary gland and the position of umbilical-cord attachment. The uterine horn is cannulated with a glass tube and ligated near to the uterine body, preventing fluid passing into the body of the uterus. Fluid is then flushed, by means of a blunt-ended needle and attached syringe, from the Fallopian tube towards the uterine horn. As the fluid passes, it takes with it any embryos present and exits via the glass cannula to be collected in a warm collecting vessel (Allen and Rowson, 1975; Allen *et al.*, 1977; Castleberry *et al.*, 1980; Imel *et al.*, 1981). Surgical recovery only allows embryos within the Fallopian tube to be recovered, i.e. those younger than 5 days. Recovery rates of the order of 70–77% have been reported (Allen and Rowson, 1975).

21.5.2. Non-surgical recovery

Non-surgical embryo recovery was first used in horses with any consistent success in Texas in 1979 (Douglas, 1979; Vogelsang *et al.*, 1979), though several other researchers had attempted it previously (Oguri and

Tsutsumi, 1972; Allen and Rowson, 1975; Douglas, 1980). It has become increasingly popular. There is no need for a general anaesthetic and hence the procedure carries lower risks. Recovery rates are reported to be as good as, if not better than, those for surgical recovery, though it does depend upon the stage of embryo development at collection. Recovery rates of up to 80% have been reported for the collection of 6–7-day-old embryos (Oguri and Tsutsumi, 1980). Collection of embryos younger than 5 days old is not possible, as this technique, unlike surgical recovery, cannot flush the mare's Fallopian tubes; hence only embryos that are within the uterine horn can be collected.

The techniques for non-surgical recovery are very similar to those used in cattle. The mare is restrained in stocks, having been prepared and washed as for minimal-contamination natural covering (see Chapter 13). A three-way catheter (French or Rusch Foley catheter) is introduced through the cervix of the mare, guided *per rectum* or by inserting a hand into the vagina and guiding the catheter through the cervix using the index finger (Fig. 21.1).

The catheter is passed as high up into the uterine horn as possible without undue pressure. Once in position, the cuff of the catheter is inflated with 15–50 ml of air via the inlet tube, so occluding the base of the uterine horn and thereby preventing the escape of flushing medium through the uterus. Fluid is then flushed in through the entry catheter up into the top of the uterine horn, and returns, along with any embryos present, via an opening into the outlet tube for collection in a warm collecting vessel (Fig. 21.2). Both horns may be flushed out simultaneously if the inflated cuff is situated within the body of the uterus (Imel *et al.*, 1980), or independently if the cuff is placed in turn in each horn (Douglas, 1979). Recovery of ova can be improved by palpation of the uterus *per rectum* at the same time as flushing (Squires and Seidel, 1995).

Both methods of recovery have their advantages and disadvantages. Surgical recovery allows younger (prior to 5 days) and therefore more robust embryos to be collected, but at a greater risk to the mare. Non-surgical

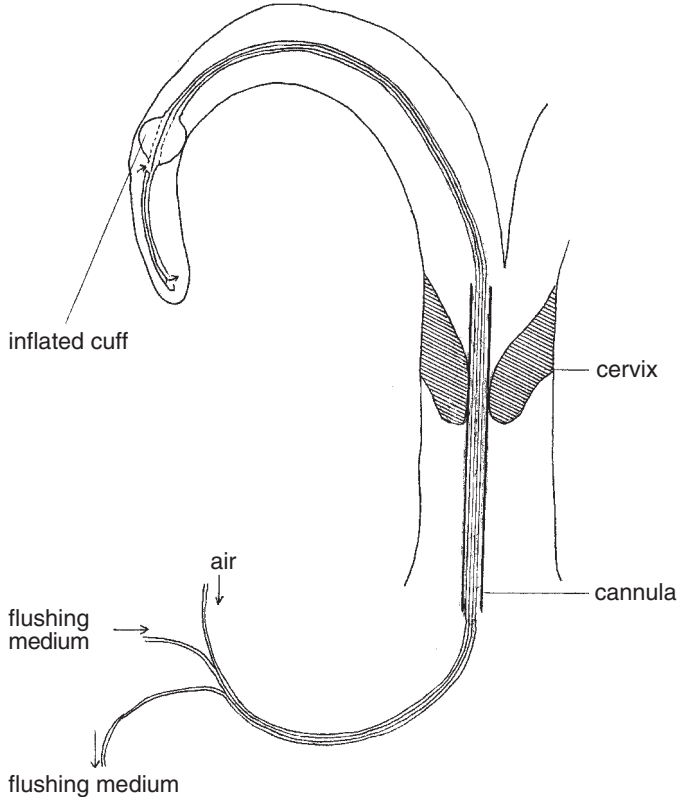


Fig. 21.1. A diagrammatic representation of the Foley catheter, illustrating the inlet and outlet tubes plus the air inlet for inflating the cuff.

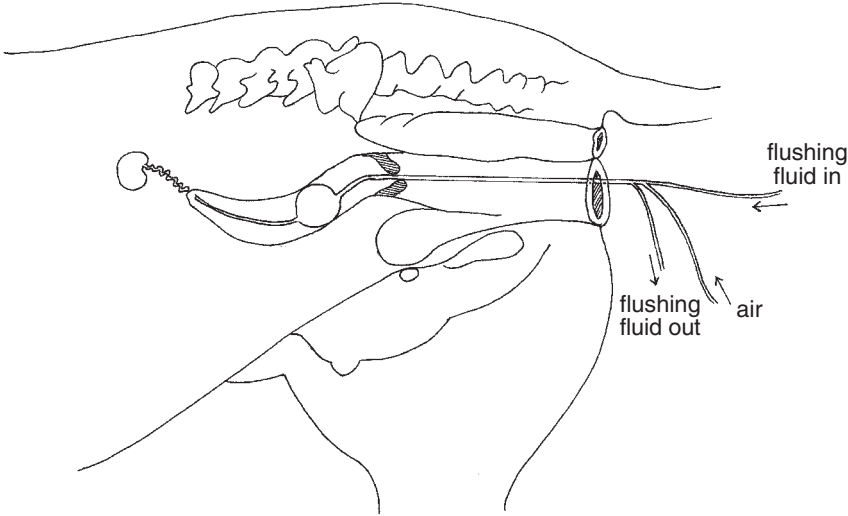


Fig. 21.2. The Foley catheter in place ready for flushing and the collection of embryos.

recovery results in older (day 6 onwards) and therefore less robust embryos, but with less risk to the mare and with potential for multiple recoveries (Iuliano *et al.*, 1985). In non-surgical recovery, the administration of PGF_{2α} at the time of collection also allows another crop of embryos to be collected from that mare within 8–9 days (Griffin *et al.*, 1981). As such, non-surgical recovery is normally the commercial method of choice.

21.6. Embryo Evaluation

Warmed, phosphate-buffered saline (PBS) or Dulbecco's fluid is normally used as the flushing medium, with the possible addition of fetal calf serum. Penicillin may also be added to guard against infection. In surgical recovery, a total of up to 50 ml can be flushed and collected. In non-surgical recovery, each donor is normally flushed twice with approximately 500 ml of fluid per flush for ponies and up to 1000 ml for horses. These flushings are then examined microscopically for embryos; a filter system may be used to aid the search. Due to their relative weight, viable embryos will sink to the bottom of the collecting vessel allowing a significant amount of the medium to be decanted off and so ease identification. Throughout the evaluation process, it is essential that embryos are kept warm (35–38°C) and that all equipment used is also prewarmed. Embryos are evaluated to assess their viability prior to transfer. They are measured and their stage of development matched to their age. Morphological features, such as shape, colour, number and compactness of cells, etc., are noted. Based upon this information, embryos are graded 1–5, 1 being excellent and 5 dead (Betteridge, 1989). Embryos graded 3 or better are normally selected for transfer (Oguri and Tsutsumi, 1972; Allen and Rowson, 1977).

21.7. Embryo Storage

Recovered embryos are either transferred immediately or chilled or frozen for use at a later date. Equine embryos can be stored for

up to 24 h at 42°C – that is, slightly above body temperature – prior to transfer, allowing some limited transportation of embryos. Embryos have been successfully transported in ligated rabbit oviducts, provided that they are transferred into recipients within 48 h (Allen *et al.*, 1976b). Alternatively, and more normally, they can be stored either in physiological saline containing 2% gelatin, in a 1 : 1 mix of mare's serum : Ringer's solution or in modified Dulbecco's PBS (Whittingham, 1971; Oguri and Tsutsumi, 1972, 1974).

Cooling embryos is a successful means of storage for 36 h at 4–5°C (Martin *et al.*, 1991). Storage mediums include modified Ham F10 medium. Commercially, embryos can be cooled and stored in an Equitainer (see Chapter 20), which acts like a cool box, cooling embryos by –3°C min^{–1} down to 5°C, and maintains them at this temperature for in excess of 48 h, allowing reasonable scope for transportation (Allen *et al.*, 1976a; Douglas-Hamilton *et al.*, 1984; Carnevale *et al.*, 1987).

The only means of long-term storage is cryopreservation (freezing). This is as yet relatively unsuccessful in horses, although quite successful and commercially viable in the sheep, cow and goat. The first successful birth of a foal from a frozen embryo was not achieved until 1982 (Yamamoto *et al.*, 1982). Success was limited, with only one live foal from 14 embryos. Czlonkowska *et al.* (1985) reported similar results. Better success rates have been achieved since – in the region of 20–40% (Slade *et al.*, 1985; Farinasso *et al.*, 1989; Squires *et al.*, 1989). Poor success rates have been postulated to be due to a variation in embryo size at freezing. Most of the embryos recovered from mares are via the non-surgical technique, hence 6–7 days old and so 500–1000 µm in diameter. Experiments have shown that cryopreservation of equine embryos larger than 250 µm in diameter gives relatively poor results. This is confirmed by Skidmore *et al.* (1990), who achieved pregnancy rates of 40–55% from frozen embryos that were less than 225 µm in diameter. It appears, therefore, that the success rates for the transfer of equine morulae and early blastocysts is quite acceptable, but the recovery rate for such

young embryos from the mare via the non-surgical technique is low compared with that for the older and larger 6–7-day-old blastocysts (Boyle *et al.*, 1989).

In order to cryopreserve embryos, a cryoprotectant, typically glycerol, needs to be used. Other cryoprotectants have been tried, with limited success (Hochi *et al.*, 1994; Ferreira *et al.*, 1997; Huhtinen *et al.*, 1997). Embryos need to be frozen in a stepwise fashion. The temperature is initially dropped down quite rapidly to -6 or -7°C and then more gradually dropped further to -33 to -35°C , followed by plunging into liquid nitrogen for storage. Storage in liquid nitrogen is presumed to be indefinite (Skidmore *et al.*, 1990). Prior to transfer, they need to be thawed out by a gradual stepwise increase in temperature, with the possible addition of sucrose to aid rehydration and help prevent alterations in osmotic pressure (Hochi *et al.*, 1996; Young *et al.*, 1997).

21.8. Embryo Transfer

The transfer of embryos can, as with collection, be done either surgically or non-surgically.

21.8.1. Surgical embryo transfer

Surgical transfer is not popular today. As with embryo collection, it requires a general anaesthetic. The mare is prepared as for surgical collection and a similar but smaller ventral midline incision is made. Just the uterine horn is exteriorized through this incision and a small hole is made at the top of the horn with a blunt needle. A Pasteur pipette or equivalent, containing the embryo, held between two bubbles of air, is introduced through this hole into the uterus and the embryo expelled into the lumen of the uterine horn. A similar, but less traumatic, procedure can be carried out through the flank of the mare while standing restrained in stocks, using a local anaesthetic and general tranquillizer. A small incision is made in the mare's flank, the uterine horn is then exteriorized through

this incision and the embryo replaced as described above.

Both these methods have the advantage that younger embryos – 2–4 days old – may be replaced into the Fallopian tubes, as access into the top of the uterine horns and the Fallopian tubes is possible. The reported success rates are very variable – 50–80% – though they do tend to be higher than those in non-surgical transfer, especially with younger, less well-developed embryos after cryopreservation (Allen and Rowson, 1975; Imel *et al.*, 1981). Pregnancy rates with cooled embryos are equivalent to fresh transfer (Sertich *et al.*, 1988; Carney *et al.*, 1991).

21.8.2. Non-surgical embryo transfer

The non-surgical transfer technique is very similar to that used in cattle and in AI (Meira and Henry, 1991; see Chapter 20, Figs 21.3 and 20.10). The mare is restrained within stocks, the perineal area is thoroughly washed and the transfer gun or catheter with attached syringe, containing the embryo held between two bubbles of air, is passed in through the mare's cervix and into the uterine body. Once in place, the embryo and the associated fluids are expelled into the uterus by slowly depressing the plunger of the syringe (Lagreaux and Palmer, 1989; Fig. 21.3).

Non-surgical transfer is a relatively easy and quick procedure. However, it does run the increased risk of introducing low-grade infections into the reproductive tract and only allows the deposition of embryos into the uterine body or the lower part of the uterine horns. This technique is therefore of limited use with embryos less than 5 days old; for 6–8-day-old embryos, pregnancy rates of 40–70% have been reported (Allen and Rowson, 1975; Castleberry *et al.*, 1980; Oguri and Tsutsumi, 1980; Wilson *et al.*, 1987).

The site of ova deposition in relation to the functional corpora lutea seems to be less crucial in the mare than in the ewe and the cow. This is presumably due to the natural transuterine migration of equine embryos. The medium in which the embryos are transferred seems to have an effect on pregnancy

Fig. 21.3. Non-surgical transfer of embryos into a recipient mare.

rates, Dulbecco's PBS being reported to give better results as a transport medium than medium 199 (Douglas, 1980).

21.9. Conclusion

ET in horses has significant potential for development, especially in the area of cryo-preservation, without which its successful commercial application is limited. At present, many breed societies refuse to accept the progeny of ET, or only allow one foal per mare per year to be registered (Bailey *et al.*, 1995). The expansion of ET within the equine industry is dependent upon several factors,

including a change in breed registration restrictions; the value of horses; the performance of ET foals; the cost of the procedures; refinement of techniques; and the attitude of the equine industry to its application. However, even within these constraints, ET in horses is a valuable experimental tool. ET may find further application and commercial worth if a reliable superovulating agent can be found and fetal sexing becomes reliable (Pieppo *et al.*, 1995; Jafer and Flint, 1996; Manz *et al.*, 1998). The alternative to superovulation, embryo bisection, resulting in identical twins, is relatively unsuccessful, with success rates at present being only of the order of 10–20%.

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